PROPHYLAXIS OF PATENT DUCTUS ARTERIOSUS WITH IBUPROFEN SUSPENSION IN PREMATURE INFANT

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PROPHYLAXIS OF PATENT DUCTUS ARTERIOSUS WITH IBUPROFEN SUSPENSION IN PREMATURE INFANTS.

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ABSTRACT

The aims of this study were to evaluate the efficacy of ibuprofen suspension in patent ductus arteriosus (PDA) prophylaxis and to determine the pharmacokinetic parameters of ibuprofen suspension in premature infants. During the ten-month-duration of the study, forty-two babies who fit our inclusion criteria were randomized to the ibuprofen (n = 22) or control group (n = 20) with a mean gestational age (GA) and birth weight (BW) of 30.64 ± 1.76 weeks, 1279.64 ± 80.33 grams and 30.20 ± 2.14 weeks, 1214.50 ± 217.52 grams, respectively. Clinical evaluation, echocardiogram and laboratory screening were performed in both groups. Ibuprofen (10 mg/kg body weight) and placebo suspension were given to the study and control groups respectively. Blood samples were drawn for serum ibuprofen level determination before the drug administration, at 2, 4, 8, 10, 12, 14 and 18 hours after the first dose, 30 minutes before and 8 hours after the second and third dose. Echocardiogram was performed at day 3, 7, 14, 21 and 28. The incidence of PDA at day 3 in the ibuprofen group was significantly lower than the one in the control group (95.45% vs 65.00%; p=0.018). There were no significant differences in renal, hematological and gastrointestinal side effects between the two groups, although the gastrointestinal irritation tended to be higher in the ibuprofen group. The pharmacokinetic parameters in the ibuprofen group were calculated from 17 subjects by using the Win Nonlin® program. There were large interindividual variabilities for Vd/F (0.3175 l/kg), Ka (0.2035 hr^{-1}), Ke (0.0244 hr^{-1}), AUC (1808.20 l/hr), T1/2 (28.42 hrs), t_{max} (10.89 hrs), and C_{max} (31.73 mcg/ml). There were no significant differences between the observed and predicted concentrations. The sustained C_{max} level above 31.73 mcg/ml after the first dose might be responsible for low PDA incidence in the ibuprofen group (95.45%); however, the exact level of ibuprofen for PDA closure needs more study. In conclusion, preterm infants who received ibuprofen suspension for PDA prophylaxis had a statistically significantly lower incidence of PDA than ones who received a placebo.

KEY WORDS: PATENT DUCTUS ARTERIOSUS/ PRETERM NEONATE/ ORAL IBUPROFEN.
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การใช้ยาน้ำแขวนตะกอนไอบูโพรเฟนในการป้องกันภาวะการเปิดของหลอดเลือดดัคตัส อาร์เตริ-โอสัส ในทารกแรกคลอดก่อนกำหนด (PROPHYLAXIS OF PATENT DUCTUS ARTERIOSUS WITH IBUPROFEN SUSPENSION IN PREMATURE INFANTS)

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บทคัดย่อ

การศึกษานี้เป็นการวิจัยประสิทธิภาพยาน้ำแขวนตะกอนไอบูโพรเฟนในการป้องกันภาวะการเปิดของหลอดเลือดดัคตัส อาร์เตริโอสัส รวมถึงหาค่าพารามิเตอร์ทางเภสัชจลนศาสตร์ของยาในทารกแรกคลอดก่อนกำหนดที่มีน้ำหนักตัวน้อย จากระยะเวลาการเก็บข้อมูล 10 เดือน มีทารกแรกคลอดก่อนกำหนด 42 คนที่เข้าการศึกษาวิจัยนี้โดย 22 คนถูกสุ่มเข้ากลุ่มที่ได้รับยาไอบูโพรเฟน มีอายุครรภ์เฉลี่ย 30.64±1.76 สัปดาห์และน้ำหนักแรกคลอดเฉลี่ย 1279.64±80.33 กรัม อีก 20 คนที่เหลือถูกจัดเข้ากลุ่มที่ได้รับยาหลอก มีอายุครรภ์เฉลี่ย 30.20±2.14 สัปดาห์และน้ำหนักแรกคลอดเฉลี่ย 1214.50±217.52 กรัม หลังได้รับยาครบ 3 วัน กลุ่มที่ได้รับไอบูโพรเฟน สามารถลดอุบัติการณ์ symptomatic PDAในกลุ่มที่ได้รับยามากกว่ากลุ่มยาหลอกอย่างมีนัยสำคัญทางสถิติ (95.45% vs 65.00%; p=0.018) และยังมีแนวโน้มลดอุบัติการณ์ของ silent PDA ได้อย่างมีนัยสำคัญทางสถิติด้วย (93.3% vs 50.0%; p=0.014) โดยไม่พบผลข้างเคียงต่อไตและระบบเลือดอย่างมีนัยสำคัญทางสถิติ แม้ว่าผลข้างเคียงต่อระบบทางเดินอาหารมีแนวโน้มเกิดในกลุ่มที่ได้รับยาไอบูโพรเฟนมากกว่ายาหลอกแต่ไม่มีความแตกต่างอย่างมีนัยสำคัญทางสถิติ จากการประเมินค่าพารามิเตอร์ทางเภสัชจลนศาสตร์ในกลุ่มที่ได้รับยาไอบูโพรเฟนจำนวน 17 คน พบความแตกต่างระหว่างบุคคลอย่างมากโดย Vd/F 0.3175 l/kg Ka 0.2035 hr^{-1} Ke 0.0244 hr^{-1} AUC 1808.20 l/hr $t_{1/2}$ 28.42 hrs t_{max} 10.89 hrs และ C_{max} 31.73 mcg/ml ไม่พบความแตกต่างอย่างมีนัยสำคัญทางสถิติระหว่างระดับยาที่วัดได้จริงกับที่ทำนายจากโปรแกรม Win Nonlin® การคงระดับยาสูงสุดในเลือด 31.73 mcg/ml หลังได้รับยาในวันแรกได้นานพออาจพบอุบัติการณ์ภาวะ symptomatic PDAต่ำกว่ากลุ่มที่ได้ยาหลอกแต่ยังต้องการการศึกษาที่มีจำนวนประชากรมากกว่านี้เพื่อยืนยันถึงระดับยาที่ปิดหลอดเลือดดัคตัส อาร์เตริโอสัส ได้แน่นอน

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LIST OF ABBREVIATIONS

6-keto-$PGF_{1\alpha}$	6-keto-Prostaglandin $F_{1\alpha}$
ANP	Atrial Natriuretic Peptide
ARR	Absolute Risk Ratio
ATP	Adenosine Triphosphate
AUC	Area under the curve
BNP	B-type Atrial Peptide
BP	Blood Pressure
BPD	Bronchopulmonary Dysplasia
BUN	Blood Urea Nitrogen
BW	Birth Weight
cAMP	Cyclic Adenosine Monophosphate
CBFV	Cerebral Blood Flow Velocity
CBV	Cerebral Blood Volume
CF	Cystic Fibrosis
Cl	Chloride
CL	Clearance
CL/F	Apparent clearance
Cmax	Maximun concentration
CO_2	Carbondioxide
COX-1	Cyclooxygenase type 1
COX-2	Cyclooxygenase type 2
CPVL	Cystic Periventricular Leucomalacia
CrCl	Creatinine Clearance
EP_1	E-prostanoid receptor type 1
F	Bioavailability
FFP	Fresh Frozen Plasma
GA	Gestational Age
GI	Gastrointestinal

LIST OF ABBREVIATIONS (CONTINUED)

Hb	Hemoglobin
Hct	Hematocrit
HPLC	High Performance Liquid Chromatography
hr(s)	hour(s)
IBU	Ibuprofen
INDO	Indomethacin
inj	injection
INR	International Normalized Ratio
IVH	Intraventricular Hemorrhage
K	Potassium
kg	kilogram
LA:A_o	Left Atrial to Aortic ratio
LAE	Left Atrial Enlargement
LPA	Left Pulmonary Artery
LV	Left Ventricle
LVE	left Ventricular Enlargement
LVO	Left Ventricular Output
MAP	Mean Arterial Pressure
mcg, µg	microgram
min(s)	minute(s)
ml	milliliter
Na	Sodium
NEC	Necrotizing Enterocolitis
no.	number
NSAIDs	Nonsteroidal Anti-inflammatory Drugs
PAP	Pulmonary Arterial Pressure
PDA	Patent Ductus Arteriosus
PGE_2	Prostaglandin E_2
PGI_2	Prostacyclin

LIST OF ABBREVIATIONS (CONTINUED)

Plt	Platelet
PO_2	Partial pressure of oxygen
PPHN	Persistent Pulmonary Hypertension of the Newborn
PRC	Packed Red Cell
PT	Prothrombin Time
PTT	Partial Thromboplastin Time
PVL	Periventricular Leucomalacia
RBC	Red Blood Cell
RCT	Randomized Controlled Trial
RD	Risk Difference
ROP	Retinopathy of Prematurity
RR	Relative Risk
Scr	Serum Creatinine
sPDA	Symptomatic Patent Ductus Arteriosus
$t_{1/2}$	Elimination half-life
Tmax	Time to maximum concentration
TNF_{α}	Tumor Necrosis Factor alpha
TT	Thromboplastin Time
UGIB	Upper Gastrointestinal Bleeding
Vd	Volume of distribution
Vd/F	Apparent volume of distribution
Vd_{ss}	Volume of distribution at steady state
WBC	White Blood Cell
wk(s)	week(s)

CHAPTER 1

INTRODUCTION

Patent ductus arteriosus (PDA) is one of the common problems found in premature infants. Patent ductus arteriosus (PDA) is inversely related to birthweight. The infants whose birthweight greater than or equal to 1360 g or gestational age greater than 28 wks have the higher ductal closure rate than the older one as reported by Clyman et al (6). The very low birthweight (VLBW) infants with respiratory distress syndrome have a very high PDA incidence (8). Left to right shunting through a PDA causes many organ effects, ie. Increased bronchopulmonary dysplasia (BPD), increased necrotizing enterocolitis (NEC), increased intraventricular hemorrhage (IVH) in CNS, and renal hypoperfusion.

Since prostaglandin E_2 (PGE_2) and I_2 (PGI_2) have major role in ductal patency, inhibition of prostaglandin could reduce PDA incidence. Indomethacin has been widely used in the prophylaxis and treatment of PDA (15-18) but with many side effects, ie. Renal dysfunction, NEC, gastrointestinal hemorrhage, periventricular leukomalacia (PVL) and IVH (17-24, 88-90). Ibuprofen, a new alternative agent has been suggested by many authorities (25-27, 29-32, 83, 87). It has the same efficacy in PDA closure as indomethacin with less vasoconstrictive effects. Many studies (26-27, 30-33, 83) reported that intravenous ibuprofen (10 mg/kg followed by 5 mg/kg at 24 and 48 hours later) had the same ductal closure rate as three doses of intravenous indomethacin (0.2 mg/kg at 12 hrs interval) with higher creatinine clearance, urine volume, and lower serum creatinine (Cr) and blood urea nitrogen (BUN)(22, 27-29, 34-35). In addition, ibuprofen didn't affect the cerebral hemodynamics and may increase cerebral blood volume (35-36). Although ibuprofen had many benefits over indomethacin, the pulmonary adverse effect had been reported (38). Ibuprofen given within the first six hours after birth in three premature infants with GA less than 28 weeks had been reported to be associated with pulmonary hypertension. In comparison studies of ibuprofen with placebo (28, 39-42), it is effective in PDA reduction at day 3

with no significant differences in serum creatinine level, BPD, IVH, PVL, NEC, and gastrointestinal bleeding. Although the meta-analysis by Shah et al (72) reported the statistically significant increase in the serum creatinine levels on day 3 in the ibuprofen group as compared with the placebo group, the values were within normal limit. All of these studies were conducted with intravenous form (ibuprofen lysine) that was not widely available in many countries including Thailand.

Efficacy of ibuprofen suspension in PDA reduction has been shown in many studies (41-43, 47, 86) with no significant differences in renal function from the control group and less renal side effects when comparing with indomethacin (44-46).

The pharmacokinetics of oral ibuprofen suspension in premature infants has been studied by Sharma et al (81). There was a large interindividual variability in pharmacokinetic parameters (54-55) but a small interindividual variability of pharmacokinetics profiles in infants older than 3 months (56-58). This difference may be resulted from the immaturity process of drug biodisposition organ and the variability of physiologic factors in different age groups. The slow absorption and elimination of drug might be used to predict mean plasma concentration that induced ductal closure. The actual concentration that is necessary for ductal closure is still unclear. The maintained serum levels of intravenous ibuprofen at 116.6±54.5 mg/l and 113.6±58.2 mg/l on day 2 and day 3 respectively have been suggested to be associated with ductal closure(28, 55) but there was still no such studies for the oral regimen. We assumed that the level produced from ibuprofen suspension may be high enough for PDA closure.

Objectives

1. To assess the efficacy of ibuprofen suspension in the prevention of PDA in premature infants.

2. To study the ibuprofen pharmacokinetics in order to correlate appropriate concentration that is probably required for maintenance of ductal closure (pharmacokinetics-pharmacodynamics relationship).

3. To evaluate the safety of ibuprofen on major organs such as renal, gastrointestinal tract and liver.

Expected outcomes

1. Ibuprofen suspension can be used for effective prophylaxis of PDA with minimal or no major organs side effects.

2. To approximate the serum drug concentration needed to close PDA in preterm infants.

CHAPTER 2

LITERATURE REVIEW

Fetal circulation

Fetal circulation is different from the adult circulation. It involves four important parts; umbilical arteries, the umbilical veins, the placenta, and the three fetal shunts (8). These three shunts have the role in the communication between each part of the heart as shown in figure 1(8).

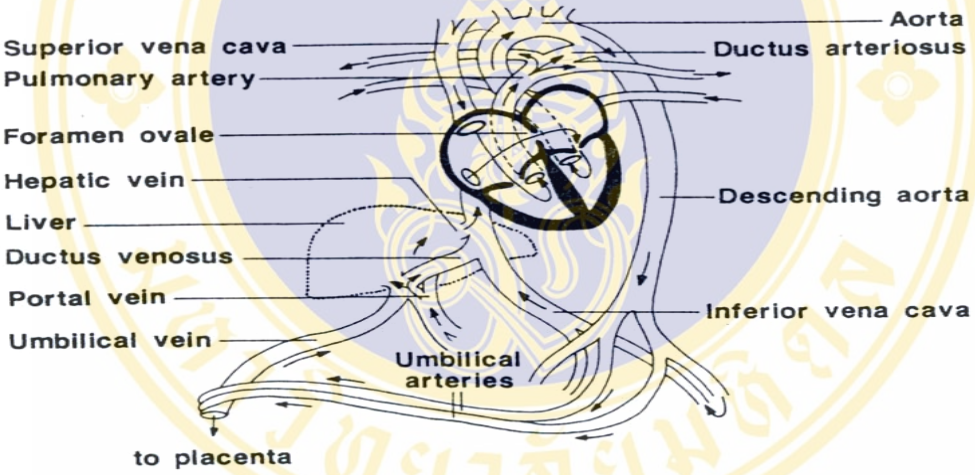

Figure 1. Fetal circulation

Firstly, the ductus venosus allows blood to bypass the hepatic circulation then enter the inferior vena cava near the right atrium. Next, the foramen ovale bypasses most of the blood from the inferior vena cava to the right side of the heart, the pulmonary circulation and enter the left atrium to the left ventricle. Lastly, the ductus arteriosus shifts the blood from the right ventricle to bypass the high-resistance pulmonary circulation and enter the descending aorta. During fetal stage, only 10% of right ventricular output flows through the pulmonary vasculature, while the remaining 90%

directly into the descending aorta via the ductus arteriosus (8). Consequently, the ductus arteriosus bypasses most of the right ventricular blood from the unexpanded lungs. It is a normal fetal vascular channel found in all mammalian fetuses, developing from the distal portion of the left sixth aortic arch and bridges the origin of the left pulmonary artery and the beginning of the descending aorta. After birth, right ventricular output must be directed into the pulmonary vasculature in order for the lung to take over the function of gas exchange as a result of the fall in pulmonary vascular resistance when the oxygenation and the lung inflation are provided by the first breaths (3). These events also occur with the elimination of low-resistance placental circulation (3) Normally, the ductus arteriosus is programmed to constrict rapidly after birth. There are three important factors dealed with the ductal closure. These factors are available muscle mass in the pulmonary vasculature, oxygen tension, and the circulating prostaglandin levels (2, 8).

In a human fetus of 4 months' gestation, the ductus arteriosus has differ histologically in aortic and pulmonary artery tissue (4). The media of the great vessels is composed primarily of elastic tissue whereas the ductus arteriosus consists mainly of muscular fibers and contains elastic tissue between only the intima and media (2,4). In this unique media, cylindrical layers of smooth muscle spiral in opposing directions, encircling the ductal channel. A prominent internal elastic lamina lies between the intima and the media and it is also distinctive, being thicker than that of the contiguous arteries at term. The intima consists primarily of an endothelial layer and loose connective tissue because of a large amount of mucoid substance in the medial wall, which is not present in other arterial tissues (2, 4). During the last trimester, intimal mounds appear on the surface of the lumen. After birth, intimal-medial dissociation begins, endothelial cells begin to separate from the internal elastic lamina, and a widened subendothelial region develops (2). This is followed by migration of smooth muscle cells into the subendothelial region, leading to the formation of mucoid cysts and nodules of muscle that appear in the inner medial layer (2). These intimal mounds continue to enlarge and eventually obliterate the ductal lumen (2). The process always begins at the pulmonary end of the ductus arteriosus and proceeds toward the aortic end (2).

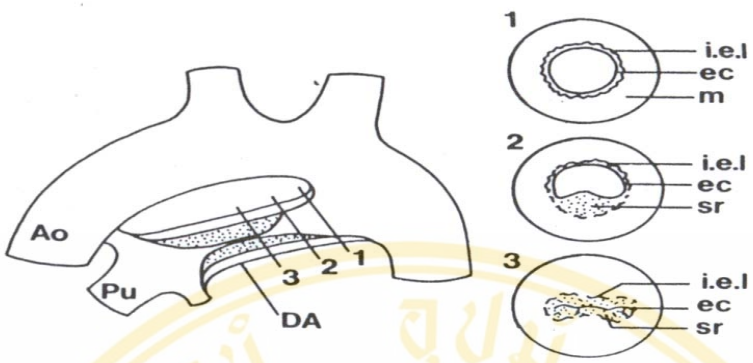

Figure 2. Schematic representation of the ductus arteriosus (taken from beagle pup) (1) The aortic end still lacks intimal thickening; (2) intimal thickening starts at the "buttom"of the ductus arteriosus; (3) closure is observed from approximately the middle to the pulmonary end. ec = endothelial cells; ie = internal elastic lamina; sr = subendothelial region; m = media. (From de Reeder EG, Gittenberger-de Groot AC, van Munsteren JC, et al: Distribution of prostacyclin synthase, 6 keto prostaglandin $F_{1\alpha}$ and 15-hydroxyprostaglandin dehydrogenase in the normal and persistent ductus arteriosus of the dog. Am J Pathol 135:882, 1989; with permission.)

In the late stages of closure, the intimal cushions proliferate and encircle the lumen almost completely, ultimately meeting to seal the ductus completely (4). In some areas fibrous tissue is deposited to fill gaps that may persist (4). The cessation of blood flow after luminal occlusion results in further ischemia to the inner layer of the media and intima (4). The result is a fibrotic core that transforms the patent ductal structure into the ligamentem arteriosum (4).

Closure of the ductus arteriosus takes place in two stages, functional and anatomic phenomenon. The first stage is the functional closure occurs during the first day or the second day of life, but the potential to reopen may remain for the next 7 or 8 days. In term infants, 20% of ducts are closed functionally by 24 hours, 82% by 48 hours, and 100% closed functionally by 96 hours; however, premature infants beyond the second week of life may show sign of PDA if PDA had not been diagnosed during the first 14 days (7). In addition, closure of PDA beyond the second weeks may not improve the infant's respiratory status (7). The second stage is the anatomical closure occurs. The proliferation of intimal cushions together with the encircling of the lumen

seal the ductus completely. The lack of blood flow after luminal occlusion causes the ischemic zone to the inner layers of the media and intima (4). Consequently, these structures transform into the ligamentem arteriosum (7).

Increased oxygen tension is considered as a potent stimulator of smooth muscle constriction. During fetal life, the partial pressure of oxygen (PO_2) to which the ductus arteriosus is normally exposed is between 18 and 28 mmHg (4). An increase in PO_2, as occurs with ventilation after birth, constricts the ductus arteriosus in mature fetal animals; however, the ductus arteriosus is not constricted by increased oxygen even at high concentrations at about 0.6 gestation (term = 150 days) (4). With advancing gestation, the amount of constriction in response to increasing PO_2 is greater and the level of PO_2 required for response falls. The mechanism of oxygen that caused the ductal closure may begin with an interaction between oxygen and cytochrome a_3 (hypothesized as the oxygen sensor) (4). It is suggested that when cytochrome a_3 becomes oxidized, ATP is generated, causing the ductus to contract, perhaps through steps linked to the control of intracellular calcium so it provides the signal for the synthesis of the constrictor endothelin-1(4).

Prostaglandins have been involved intimately in mediating both prenatal and postnatal ductal closure. PGE_2 is thought to be the major mediator of ductal relaxation. Many reports have suggested that preterm infants with patent ductus arteriosus and respiratory distress syndrome have significantly higher levels of PGE_2 than do normal preterm infants of the same age. Clyman et al. (6) showed that the PGE_2 concentrations during the first two postnatal days were higher (17.10 ± 3.10 pg/ml plasma ± SEM, $n = 23$) than they were after the third postnatal day (6.12 ± 0.63 pg/ml, $n = 19$, $p<0.005$). By the third postnatal day, however, arterial concentrations were similar to adult venous concentrations (7.50 ± 0.78 pg/ml, $n = 5$). There was no significant difference between the plasma PGE_2 concentrations in the 12 infants with PDA (measured at a time when they were exhibiting signs of a left-to-right shunt) and the concentrations in the 17 infants who never had signs of a PDA during their hospitalization. These findings do not support the hypothesis that an increased concentration of circulating PGE_2 is the primary cause for the persistent patency of the ductus arteriosus in preterm infants with respiratory distress

syndrome(6). Therefore, PGE_2 would have to be produced locally in the wall of the ductus if it were to be primarily responsible for ductal patency. Moreover, production of PGE_2 is inhibited at the low ambient oxygen tensions associated with persistent patency of the ductus arteriosus (6).

Prostacyclin (PGI_2), another prostanoid vasodilator, is the major arachidonic acid product of the ductus arteriosus. Although it is significantly less potent than PGE_2, it is much more prevalent. It is produced at levels about 10 times those of PGE_2 especially in smooth muscle cells at the sites of intimal thickening than at other sites. In contrast to PGE_2, PGI_2 production has been shown to increase under the hypoxic conditions known to be associated with patency of the ductus. The production of 6-keto-$PGF_{1\alpha}$ (a prostacyclin metabolite) under hypoxic condition was decreased and production was increased with hyperoxia. In vitro, the ability of PGI_2 to dilate the ductus is two to three orders of magnitude less than that of PGE_2.

The role of cyclooxygenase (COX) and E-prostanoid receptors in regulating ductus arteriosus tone were also studied (68). The expression of COX isoenzymes to the regulation of ductus arteriosus tone has been reported. COX-2 expression plays a major role in maintaining fetal ductus arteriosus during late gestation (immediately following birth); however, COX-1 plays an important role in ductus arteriosus tone (during fetal stage) possibly via the generation of peripheral (nonductal) prostaglandins. Indeed, COX-1 inhibition markedly reduces levels of PGE_2 in the plasma and causes a significant constriction of the ductus arteriosus (68). As a result, the selective COX-2 inhibitor may be safer for the fetus than nonselective COX inhibitor although the precise risks of perinatal COX-2 inhibition remain unknown (68).

Once the COX isoenzymes have yielded PGE_2, the prostaglandin exerts its effects on ductus arteriosus through the major E-prostanoid (EP) receptors (EP_1, EP_2, EP_3, and EP_4) (68). Activation of EP_1 increases IP_3 formation and elicits vasoconstriction, whereas stimulation of EP_2 and EP_4 increases cyclic adenosine monophosphate (cAMP) levels and leads to vasodilation; stimulation of EP_3 decreases cAMP but acts as vasodilator and evokes ductus arteriosus relaxation through a process dependent on potassium channels that are sensitive to adenosine triphosphate (ATP). However, EP receptor subtypes 2 to 4 are expressed equivalently in fetal

ductus arteriosus relaxation. EP_4 receptor may be suggested to represent a feasible target for the treatment of PDA in premature neonates and a selective EP_2 agonist might be effective in maintaining DA patency in ductus-dependent circulation such as congenital heart disease (68).

The prostaglandin and oxygen systems do not function independently, but they interwined on many levels include the metabolism, the pathophysiology, and in the clinical setting. In addition to the influence of hypoxia on prostaglandin synthesis, there is an interaction at the level of end organ responsiveness. In conclusion, normal spontaneous ductal closure occurs as the relation of the following pathophysiologic mechanisms (4). First, the smooth muscle contracts directly in response to the postnatal increase in PO2. Second, the level of PGE2 decreases as a result of decreased synthesis or increased catabolism of this mediator. Next, the response of ductal tissue to the prostaglandin decreases while the contractile effects of ductal tissue to oxygen increases with advancing
age. Last, the PGE2 and oxygen act synergistically at the biosynthesis level and/or organ response.

Pathophysiology of PDA

The incidence of PDA is inversely related to birthweight. The patients who demonstrated spontaneous closure of the ductus arteriosus had higher mean birth weight than the patients who required either medical or surgical closure of the ductus arteriosus. The patients whose birthweight are greater than or equal to 1,360 g have higher ductal closure rate than the others (6). Additionally, the patients whose gestational age is greater than 28 weeks had more percent ductus closed (6).

With continued patency there is a progressive increase in the volume of blood shunted via the PDA to the pulmonary circuit so the clinical manifestations and laboratory data can be summarized as following table (6).

Table1. Features of significant PDA in a premature infant with respiratory distress syndrome

Clinical findings Continuous or systolic murmur Hyperactive left ventricular impulse Wide pulse pressure (>35 mmHg) Bounding peripheral pulses Tachycardia Tachypnea Enlarged liver Edema
Laboratory data Enlarged heart by roentgenogram Enlarged left atrium and left ventricle by echocardiography PDA diagnosed by Doppler, contrast echo or other imaging technique
Ventilatory requirements Prolonged IMV/CPAP (intermittent mandatory ventilation / continuous positive airway pressure) Reinstitution of IMV/CPAP Increasing level of ventilatory support

Another report by Bhat et al. (8) supported the direct relation between the incidence of patent ductus arteriosus and gestational age. In preterm infants with a gestational age less than 32 weeks who do not develop hyaline membrane disease, symptomatic patent ductus arteriosus generally becomes evident between two and seven days of age (a full spectrum of clinical signs by about 96 hours of age) (8). On the contrary, in premature infants with hyaline membrane disease, symptomatic patent ductus arteriosus appears at 72 to 96 hours after birth (typically show signs of a left-to-right shunt at 48 to 72 hours), presenting as an increase in oxygen requirement and prolongation of ventilator dependence, which indicates increased pulmonary complications (8).

Additionally, symptomatic PDA is associated with increased mortality and morbidity, including bronchopulmonary dysplasia, necrotizing enterocolitis, feeding intolerance, and intracranial hemorrhage. We can summarize the incidence and the pathophysiology of PDA at different birth weight as following table.

Table2. Different faces of patent ductus arteriosus.

	VLBW (<1000g)	LBW(1201-2000g)	Full term (>2000g)
Incidence	80%	40%	1/2500-5000
Clinical manifestations	Respiratory distress syndrome	*Respiratory* -Failure to wean -Increased O_2 requirement *Cardiac* -Bounding pulses -Murmur -CHF *General* -Failure to thrive	Often asymptomatic Murmur CHF SBE
Onset of symptoms	From birth	2d – 3wk	6 – 8wk
Associated factors	Severe prematurity	-RDS -Hypoxia -Hypocalcemia -Fluid overload	-Other congenital heart malformations -Chromosomal or viral syndromes
Therapy	Prophylactic indomethacin	-Indomethacin -Surgery	-Antibiotic prophylaxis for surgical procedures -Surgical ligation
Sequelae	-Increased BPD -Increased NEC -Renal hypoperfusion -Increased IVH		-Pulmonary hypertension -Infective endocarditis
CHF = congestive heart failure; SBE = subacute bacterial endocarditis.			

Study by Skinner (69) suggested that clinical signs alone cannot be absolutely indicated left-to-right ductal shunting in the preterm. The presence of classical physical signs above means that there usually (but not always) presences a big left-to-right shunt, but their absence does not exclude a large shunt, especially in the first 2 days and in extreme prematurity. However, the classical features of cardiac failure are a useful guide in larger infants and those more than 3 to 4 days old. Because of the

higher incidence of ductal patency in very preterm neonates, echocardiographic examination should be performed in order to prove PDA or silent PDA as following six criteria.

1. Bowing of the interatrial septum to the right with enlarged left atrium and left ventricle - four chamber views.

2. Left atrium enlarged; LA : A_o ratio > 1.4:1 – long axis view.

3. Colour Doppler-continuous flare in the main pulmonary artery from arterial duct.

4. Pulsed wave Doppler-turbulent flow in the main pulmonary artery, continuous antegrade flow in diastole in left pulmonary artery and arch of aorta, retrograde diastolic flow in descending aorta, cerebral and gut blood vessels.

5. Continuous wave Doppler-continuous left-to-right flow in main pulmonary artery from the ductus, with low velocity (<1m/sec) at end diastole.

6. Raised left ventricular stroke volume.

To categorize the size of the shunt, Table 3 is useful to list the finding.

Table3. Comparison of the degree of shunting examined by echocardiogram.

Echocardiographic feature	Small left-to-right shunt	Moderate left-to-right shunt	Large left-to-right shunt
Ductal size on color	<1.5mm	1.5-2.0mm	>2mm
LA:A_o ratio*	<1.4:1	1.4:1-1.6:1	1.6:1
Diastolic flow in descending aorta	Mostly antegrade down to zero	Zero or modest reversal (<30% of forward flow)	Reversal throughout diastole (>30% forward flow)
Anterograde peak diastole flow velocity in LPA	<30cm/s	30-50cm/s	>50cm/s

* Expect lower values when the atrial septal defect (foramen ovale) is wider than 2 mm and with fluid restriction (which is the norm in many units)

To differentiate other features of a large left-to-right shunt, table 4 may be helpful as a “rule of thumb” guide (69).

Table 4. Comparison of the level of myocardial failure by echocardiogram.

Clinical/echocardiographic feature	No/minimal myocardial failure	Moderate myocardial failure	Severe myocardial failure
LV functional shortening (%)	>40%	30-40%	<30%
LV stroke volume (ml/kg)	>2.5	1.5-2.5	<1.5
LV output (ml/kg/min)	Markedly elevated (>350)	Mildly elevated (200-350)	Not elevated (<200)

Because haemodynamically significant PDA worsens respiratory distress by initiating pulmonary congestion and inactivating surfactant, an increase in natriuretic hormone levels in the circulation can be initiated. Additionally, high ductal flow from the decreasing of pulmonary vascular resistance can activate many mechanisms for natriuretic hormone secretion such as the dilation of left atrium and the activation of the sympathetic nerve system as well as the renin-aldosterone-angiotensin system. The study by Pesonen (10) suggested that atrial natriuretic peptide (ANP) and B-type atrial peptide (BNP) can be used to predict ductal size such as ductal flow. Even if there is large interindividual variation in hormone levels with similar ductal flow, the hormone concentrations might be useful in the follow-up of open ductus patients because of reproducible and rapid results of BNP concentration (10). Using the cut-off points of 5000 pmol/l for Nt-pro ANP (the much more stable form of ANP) and 25 pmol/l for BNP is the useful clinical guide to estimate the ductal size.

In addition to respiratory distress syndrome caused by PDA, left-to-right shunting through the duct is associated with the occurrence of cystic periventricular leucomalacia (CPVL). Pladys et al. (11) compared 12 infants who developed CPVL with those who did not. On day 1, symptomatic PDA was more frequent (64% vs

26%; p=0.03) in the CPVL group, left ventricular output was higher (median = 341 vs 279 ml/kg/min; p=0.005), and rescue surfactant was more frequently used (83% vs 47%; p=0.03). Consequently, the occurrence of CPVL could lead to the presence of symptomatic PDA.

Phototherapy and ductal reopening

Phototherapy is frequently used for the treatment of neonatal hyperbilirubinemia especially in premature infants. Though phototherapy usually controls non-haemolytic hyperbilirubinemia successfully, many side effects are known; ie. changes in body temperature, peripheral blood flow, gastrointestinal tract motility, electrolyte and water imbalance and insensible water loss. Additionally, phototherapy, generally begun on the first day of life, is frequently given to patients with PDA. Rosenfeld et al. (12) reported the relationship between phototherapy and the incidence of PDA in premature infants. The incidence of PDA was significantly decreased in babies with shielding (shield 11/36 vs no shield 23/38; p=0.009). Moreover, shielded patients had shorter duration of hospital stay than nonshielded group (74 vs 85 days; p<0.05). In conclusion, shielding may be a practical method to decrease the complication of PDA in premature infants.

Benders and colleagues (13) also reported the changed in left ventricular output (LVO), ductal flow and left pulmonary artery (LPA) blood flow during phototherapy in 27 preterm infants of less than or equal to 32 weeks gestational age. More than fifty percent (14/27) of the infants, the ductus arteriosus reopened during phototherapy. In infants in whom the ductus did not reopen, LVO decreased immediately after the onset of phototherapy and was significantly lower after 30 minutes and 2 hours of exposure when compared with infants whom the ductus did reopen. After long exposure, there were no differences in LVO between the two groups (13).

Consequently, chest shielding reduced the incidence of PDA by prevention of light penetration.

Pharmacological treatment and prophylaxis for patent ductus arteriosus

Indomethacin

Indomethacin has gain wide acceptance as a potent constrictor of the ductus arteriosus in preterm infants with respiratory distress syndrome. Over the years, indomethacin therapy has been shown to be successful in mediating ductal closure in 70% to 90% of treated neonates and become accepted as being quite effective in the premature infant. Because prostaglandins had a major role on the pathophysiology of PDA, indomethacin (cyclooxygenase inhibitor) could reduce the level of PGE2 that caused ductal constriction. The use of indomethacin in PDA treatment was first reported in 1970 by Heymann (15) and Friedman (16).

Prophylactic indomethacin

Many studies (15-24) demonstrated the advantage and disadvantage of prophylactic indomethacin in reducing the incidence of PDA. The advantages include the decreased subsequent incidence of left to right shunting, increased rate of permanent ductal closure and decreased the incidence and severity of IVH especially grade III and grade IV IVH, pooled of risk difference -0.039 (95%CI -0.060, -0.011) (18).

Although there are sufficient data of reduction in the incidence of PDA, a recommendation to prophylactically administer indomethacin to all premature neonates remains debatable. Prophylactic indomethacin can reduce cerebral and mesenteric blood flow velocity and increased cerebral and mesenteric vascular resistance (18). Despite fewer subsequent occurrence of PDA, there is no improvement in overall respiratory morbidity or mortality, and there does seem to be a trend towards an increased incidence of necrotizing enterocolitis (18). Meta-analysis and systemic review by Fowlie (19) reported a trend in increased incidence of necrotizing enterocolitis, pooled estimate of risk difference 0.015 (95%CI -0.002, 0.033). Additionally, some treatment may transiently impair renal function.

Therapeutic indomethacin

Many studies (20) supported higher ductal closure rate in early symptomatic therapy group indicated by the reduction in morbidity, the need for surgical ligation,

and the lower incidence of NEC. Additionally, infant's birthweight can determine the chance of PDA closure rate (20). The larger infants (birth weight over 1 kg) have the higher chance of closure rate.

Continuous infusion

According to many well-known side effects from vasoconstriction induced by bolus injection of indomethacin, continuous injection was suggested as new alternative.

Christmann et al (22) compared the changes in cerebral, renal, and mesenteric blood flow velocities after continuous infusion over 36 hours versus three bolus injection of indomethacin. The result showed that during continuous infusion of indomethacin, there was no significantly change in cerebral, renal, and mesenteric blood flow velocities, whereas, the flow velocities in the infants receiving bolus injections decreased significantly during the first two hours after indomethacin administration. There was a transient, but significant reduction in urine output after bolus injection. In contrast to bolus injection, there were no decrease of organ blood flow and impairment of urine output following continuous infusion over 36 hours. Similar to the study by Hammermann et al. (21), the decrease in cerebral blood flow velocity after indomethacin injection was eliminated by administering as a continuous infusion over 36 hours.

In summary, continuous intravenous therapeutic indomethacin infusion is clearly less toxic than bolus administration.

Prolonged infusion

Normally, ductal closure occurred in two phases, functional closure followed by anatomical closure. Until full anatomical closure is achieved, vasoconstriction is reversible thus the infant remains at risk for recurrence. Conventional indomethacin therapy transiently suppressed dilator prostanoid production, facilitating ductal vasoconstriction but not always allowing sufficient time for anatomic ductal closure.

Tammela et al. (23) evaluated whether a prolonged low-dose course of indomethacin 0.1 mg/kg every 24 hours for 7 times would produce an improved closure rate and have fewer side effects compared with a short standard dosage schedule 0.2 to 0.1 to 0.1 mg/kg in 24 hours. The primary PDA closure occurred more often in the short course group (94% vs 67%; P=0.011) but the sustained closure rates

were not different (74% vs 60%). The short course group also has less frequency of surgical ligation, shorter duration of oxygen supplement, less NEC, and lower urea retention rate. As prolonged administration of low-dose indomethacin seems to be associated with less effective and higher rate of surgical ligations compared with a standard three doses course, a prolonged low-dose regimen does not offer a better choice compared with a standard short course dose in treatment of symptomatic PDA in preterm infant(14, 23).

Roque et al. (24) reported the contrast result that a prolonged low-dose protocol 0.1 mg/kg intravenous administration for six days had overall rate of primary closure 84.7% with cumulative dose 0.35 mg/kg approximately half of the 0.6 mg/kg dose used in the classic protocols. The treatment protocol was well tolerated, with minor biological disturbances and only one case showed transient oliguria. In conclusion, the prolonged low-dose indomethacin did not achieve better short or long term closure; however, a 3-dose course of the normal indomethacin followed by a prolonged maintenance phase of low dose indomethacin 0.1 mg/kg/day for 5 days may be therapeutically advantageous in
preventing recurrences of PDA (24). Additionally, the practical guidelines to the medical treatment of PDA in premature neonate can be summarize.

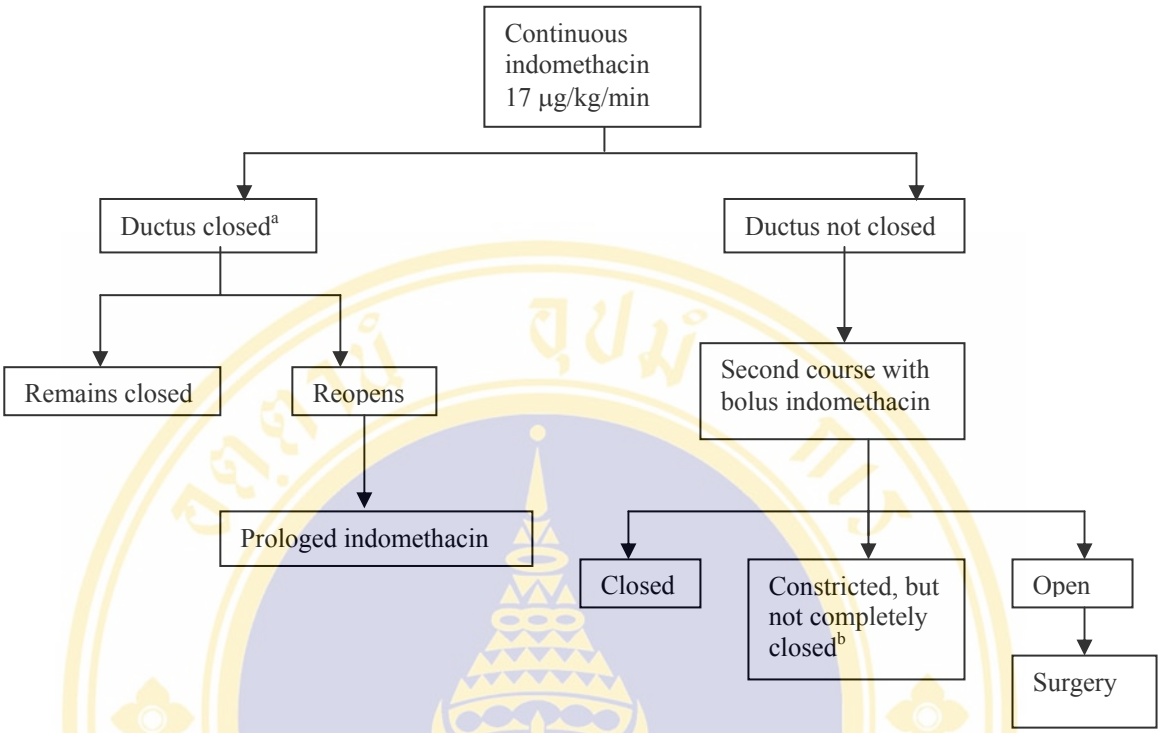

Figure 3 Therapeutic use of indomethacin for patency of the ductus arteriosus practice guidelines

a. **Evaluate no sooner than 12 hours after the completion of therapy.**
b. **Each patient must be individually evaluated. Some patients may respond to a third course at a higher dose, or to a more prolonged course; some may require surgery.**

Recently, there are five randomized controlled trials (88) comparing prolonged versus short course indomethacin differ in the dosage regimes, diagnosis of PDA, and onset of treatment. The result could be summarized as following table.

Table 5 Prolonged vs short course indomethacin in treatment of PDA in preterm infants

Citation	Study group	Study type (level of evidence)	Outcome	Key results	Comments
Rhodes et al (1988)	70 preterm infants < 1500 g with echocardiographically diagnosed PDA were randomized to either prolonged course indomethacin over 1 week or to short course (2 doses of indomethacin;n=36). All infants were given 2 doses of indomethacin 0.15 mg/kg 12 hours apart. The prolonged course group (n=34) received additional 0.1 mg/kg once daily hourly for 5 days.	Prospective randomized controlled trial (level 1b)	Closure after first course Recurrence of PDA Need for surgical ligation	RR 1.11 (95% CI 0.77 to 1.61); RD 0.06 (95% CI -0.16 to 0.29) RR 1.51 (95%CI 0.65 to 3.52);RD 0.10 (95%CI -0.10 to 0.30) RR 2.12 (95%CI 0.20 to 22.30); RD 0.03 (95%CI -0.06 to 0.13)	No blinding of intervention. No differences in mortality rates.
Hammerman and Arambura (1990)	30 infants < 1500 g with echocardiographically confirmed PDA were randomized to receive standard indomethacin therapy (0.2 mg/kg/dose 8 hourly), followed by either maintainance indomethacin (0.2 mg/kg 0nce daily for 5 days; n=20) or equivalent volume of placebo for 5 days (n=19).	Prospective randomized controlled trial (level 1b)	Closure after first course Recurrence of PDA Need for surgical ligation	RR 1.22 (95% CI 0.90 to 1.66); RD 0.16 (95% CI -0.07 to 0.40) RR 0.11 (95%CI 0.01 to 1.84);RD -0.21 (95%CI -0.41 to -0.01) RR 0.14 (95%CI 0.02 to 1.00); RD -0.32 (95%CI -0.56 to 0.08);NNT 3.0 (95%CI 2 to 12)	Double blind study. There was no increase in the toxic effects of indomethacin.
Rennie and Cooke (1991)	Total of 121 infants <2500 g with clinical signs of PDA were randomized to receive either prolonged course indomethacin (0.1 mg/kg once daily for 6 days; n=59) or short course (0.2 mg/kg 12 hourly for 3 doses; n=62).	Prospective randomized controlled trial (level 1b)	Closure after first course Recurrence of PDA Need for surgical ligation	RR 1.16 (95% CI 0.99 to 1.36); RD 0.12 (95% CI -0.01 to 0.25) RR 0.61 (95%CI 0.32 to 1.17);RD -0.12 (95%CI -0.27 to 0.03) RR 1.58 (95%CI 0.27 to 9.10); RD 0.02 (95%CI -0.05 to 0.09)	No blinding. Echocardiography was not used for assessment of PDA. Higher mortality rate in the prolonged indomethacin group, not directly related to treatment. Majority occurred after the first month.
Tammela et al (1999)	61 infants of gestational ages 24-32 wk with a PDA confirmed with echocardiography were randomized to receive short course indomethacin (3 doses of 0.2, 0.1, and 0.1 mg/kg in 24 hours; n=31) or prolonged course (0.1 mg/kg q 24 hr hourly x 7 days). Echocardiography was performed 3, 9, and 14 days after starting treatment.	Prospective randomized controlled trial (level 1b)	Closure after first course Recurrence needing treatment Need for surgical ligation	RR 0.71 (95% CI 0.54 to 0.93); RD -0.27 (95% CI -0.46 to 0.08) RR 1.03 (95%CI 0.37 to 2.85);RD 0.01 (95%CI -0.19 to 0.21) Increased need for surgical ligation in the prolonged group ;RR 4.65 (95%CI 1.09 to 19.78); RD 0.24 (95%CI 0.05 to 0.42); NNH 4.0(95%CI 2 to 20)	Only assessment was blinded. No difference in mortality rates.
Lee et al (2001)	Infants ≤ 1500 g with a symptomatic PDA greater or equal to 1.5 mm on echocardiography were randomized to conventional indomethacin (0.2 mg/kg/dose q 12 hourly x 3 doses; n=70) or prolonged low dose course indomethacin (0.1 mg/kg q 24 hourly x 6 doses; n=70).	Prospective randomized controlled trial (level 1b)	Closure after first course Need for surgical ligation	RR 1.02 (95% CI 0.87 to 1.27); RD 0.01 (95% CI -0.14 to 0.17) RR 0.62 (95%CI 0.27 to 1.39); RD -0.07 (95%CI -0.19 to 0.05)	No blinding of intervention. Intention to treat analysis. PDA diagnosis by echocardiography. No difference in mortality rates.

From the table, there is significant "in between study heterogeneity" as gestational age, birth weight, failure of PDA closure, need for surgical ligation, recurrence of PDA, and mortality rates. In the majority of studies, the incidence of renal side effects was less in the prolonged indomethacin group compared to the short course group. There were no significant differences in the other co-morbidities.

In conclusion, we can summarize the advantage and the disadvantage for each type of indomethacin administration by table 6.

Table 6 Advantages and disadvantages of the standard indomethacin administration regimens

Type of administration	Clinical advantage	Clinical concerns
Early therapeutic	-Treat only those with PDA -Best response with earliest post natal treatment	
* bolus	More convenient	Decreased renal, cerebral, and mesenteric blood flow
* continuous	Virtually eliminates reductions in blood flow	No definitive study to prove efficacy equal to bolus
Prolonged administration	Possible reduction in ductal recurrence rate	Possible increased in NEC, oxygen requirement, IVH, rise in creatinine and urea.

Knight (61) concluded that the randomized controlled trials of PDA closure could be categorized into three groups as followings.

1. Prophylactic treatment in first 24 hours after birth.

There are 14 trials that treat in the first 24 hours of life without using any assessment of ductal patency or shunt size as selection criteria.

Table 7 Prophylactic treatment in first 24 hours after birth

1st author	Year	n	Weight (g)	Inclusion criteria	Dose (mg/kg)	Interval	1st dose	Placebo
Mahony	1985	104	700-1300	-	0.2/0.1/0.1	12 then 24 h	12-18 h	Yes
Ment	1985	48	600-1250	No IVH	1st dose 0.2 or 0.1 then 0.1	12 h×5	6-9 h	Yes
Puckett	1985	25	<1400		0.2	12 h×3	<24 h	Yes
Rennie	1986	50	<1750	No IVH	0.2	24 h×3	<24 h	Yes*
Krueger	1987	32	750-1500	RDS IPPV	0.2	×1	24 h	No
Vincer	1987	30	<1500	CPAP or IPPV	0.2	12 h×3	12 h	Yes
Hanigan	1988	122	≤1500	No IVH	0.1	<12, 24, 48, 72 h old	<12 h	Yes
Ment	1988	36	600-1250	No IVH	0.1	24 h×3	6-10 h	Yes
Bandstra	1988	199	300-1300	Added O_2	0.2/0.1/0.1	12 h×3	<12 h	Yes
Bada	1989	141	≤1500	No IVH	0.2/0.1/0.1	12 h×3	6 h	Yes
Crouser	1996	90	600-1250	Surfactant	0.1	24 h×6	<24 h	Yes
Ment	1994	431	600-1250	No IVH	0.1	24 h×3	6-12 h	Yes
Ment	1994	61	600-1250	IVH	0.1	24 h×3	6-12 h	Yes
Cassidy	1989	84	≤1000	Added O_2	Ligation		<24 h	No

The only beneficial results of these trials are a reduction in IVH and severe IVH (RR 0.74, CI 0.63-0.87 for any IVH in 11 trials, RR 0.59, CI 0.41-0.84 for grade 3 and 4 IVH in 8 trials and RR 0.39, CI 0.19-0.80 for grade 4 IVH in 6 trials. Fowlie et al-- also confirm the significant reduction in rates of severe IVH in infants given indomethacin but it is not accompanies by renal side effects, GI perforation, and necrotizing enterocolitis. However, the improvement in rate of IVH in this review did not translate to improvement in rates of neurosensory impairment.

2. Pre-symptomatic treatment of PDA

Table 8 Pre-symptomatic treatment of PDA

1st author	Year	n	Birth weight	Criteria	Ductus assessment	Dose	Interval	Age onset	Placebo
Presymptomatic indomethacin									
Kaapa	1983	27	2096 mean	RDS PDA	Aortogram	0.2 oral	24 h	6-68 h	No
Hammerman	1987	24	≤1000	PDA	Contrast echo	0.2	12 h × 3	48-72 h	Yes
Vogtmann	1988	41	≤1500	PDA	Clinical or systolic time intervals	0.2 oral	12 h × 3	3rd day	No
Weesner	1987	26	<1750	RDS IPPV PDA	Asymptomatic+contrast echo	0.3	24 h × 3	<48 h	Yes
Van Overmeire	1998	86	1275 mean	RDS IPPV PDA	Echo angiogram	0.2	12 h × 3	Day 3	No
Mahony	1982	47	<1700	PDA	Asymptomatic murmur	0.2/0.1/0.1	0, 12, 36 h	70 h	Yes
Mullett	1982	47	<1750	PDA	Murmur	0.2 oral	24 h × 2	7.5 days	Yes
Symptomatic									
Cotton	1978	25	794-1361	IPPV+PDA	Hemodynamically significant		Ligation	8 days	No
Yeh	1981	55	<2040	PDA	Hemodynamically significant	0.3	24 h × 3	9.9 days	Yes
Yanagi	1981	39	1340 mean	RDS+IPPV	Hemodynamically significant	0.2 oral	8 or 24 h × 3		Yes
Merritt	1981	25	≤1350	RDS IPPV PDA	Clinical or M Mode or aortogram	0.2	24 h × 3	48 h	No
Rudd	1983	30	<1500	PDA	Hemodynamically significant	0.2 oral	24 h × 3	8 days	Yes
Gersony	1982	405	<1750	PDA	Hemodynamically significant	0.2/ligation	12 h × 3		Yes

There are no benefit in respiratory outcome in treating at a presymptomatic stage, and none in treating all babies <1000 g early. Moreover, there are no significant changes in death (RR 0.74, CI 0.38-1.46), ROP or NEC in the indomethacin treated groups but there is a significant reduction in the subsequent need to treat the ductus arteriosus (RR 0.38, CI 0.26-0.55).

3. Later treatment with haemodynamically significant PDA

As with the other group of trials, the infants randomized to indomethacin had a lower chance of needing further treatment of the ductus (RR 0.32, CI 0.23-0.44). There was no difference in the occurrence of BPD (RR 1.11, CI 0.85-1.40) or death (RR 0.86, CI 0.60-1.24). However, these trials do not give useful information on the

contribution of PDA to outcome or the need for PDA closure. Moreover, there is no improvement in outcome from treatment of the PDA soon after it becomes symptomatic.

Table 9 Pooled data of randomized trials of indomethacin and surgical ligation of PDA. Results for IVH, PDA, NEC and ROP are for indomethacin trials only.

Outcome	Relative risk	95% CI
Prophylactic treatment		
IVH all grades	0.74	0.63-0.87
IVH grade 3 or 4	0.59	0.41-0.84
IVH grade 4	0.39	0.19-0.80
Death	0.91	0.73-1.14
PDA	0.32	0.25-0.42
BPD	1.07	0.91-1.25
ROP	0.95	0.78-1.17
NEC	1.27	0.67-2.40
Pre-symptomatic treatment		
Death	0.74	0.38-1.46
PDA	0.38	0.26-0.55
BPD	0.87	0.57-1.31
ROP	0.87	0.36-2.09
NEC	0.58	0.11-3.18
Symptomatic treatment		
Death	0.86	0.60-1.24
PDA	0.32	0.23-0.44
BPD	1.11	0.85-1.44
ROP	0.35	0.12-1.03
NEC	1.02	0.49-2.13

Ibuprofen

Although indomethacin has therapeutically effective in reducing the incidence of PDA, it is known to be associated with certain adverse effects, which are predominantly mediated by the vasoconstriction. Reduction of intestinal blood flow, renal blood flow, and cerebral blood flow are common important side effects that can be found.

Ibuprofen is rapidly emerging as a potential alternative to indomethacin in the treatment of PDA. Ibuprofen appears to be effective in mediating ductal closure while possibly causing less vascular compromise. We can summarize the comparison studies of ibuprofen and indomethacin in PDA closure as following table.

Table 10 Comparison of the study of ibuprofen and indomethacin in PDA closure.

First author/Year/ Study type	Population/Gestational age	Dose of ibuprofen	Dose of indomethacin	Number of infant in which the duct closed (ibuprofen)	Number of infant in which the duct closed (indomethacin)
Mosca, 1997, RCT	< 31weeks echocardiographically proven PDA	10 mg/kg (n=8) one dose	0.2 mg/kg (n=8) one dose	5/8 (63%) after one dose 3/3 (100%) after repeat doses	6/8 (75%) after one dose 2/2 (100%) after repeat doses
Overmeire, 1997, RCT	< 33weeks RDS 2-3 days of age	10mg/kg, followed by 5mg/kg 24 and 48 hours later	0.2 mg/kg at 12 hour interval for three doses	16/20 (80%)	15/20 (75%)
Patel, 1995, RCT	23-28 weeks echocardiographically proven PDA	5 mg/kg (n=12) 10 mg/kg (n=6) one dose	0.1 mg/kg (n=15) one dose	10/18 (57%)	9/15 (57%)
Overmeire, 1998, RCT	Preterm echocardiographically proven PDA	10 mg/kg followed at 24 and 48 hour later by 5 mg/kg.	0.2 mg/kg at 12 hour intervals for three doses	39/52 (75%)	37/51 (73%)
Patel, 2000, RCT	23-35 weeks echocardiographically proven PDA	10 mg/kg followed at 24 and 48 hour later by 5 mg/kg.	0.2-0.25 mg/kg at 12 hour intervals for three doses	14/18 (78%)	14/15 (93%)
Overmeire, 2000, prospective multicenter trial	≤ 32weeks echocardiographically proven PDA	10 mg/kg followed at 24 and 48 hour later by 5 mg/kg.	0.2 mg/kg at 12 hour intervals for three doses	52/74 (70%)	49/74 (66%)
Lago, 2002, RCT	≤ 34weeks echocardiographically proven PDA	10 mg/kg followed at 24 and 48 hour later by 5 mg/kg.	0.2 mg/kg at 12 hour intervals for three doses	69/94 (73%)	56/81 (69%)
Su PH, 2003, RCT	≤ 32weeks echocardiographically proven PDA	10 mg/kg followed at 24 and 48 hour later by 5 mg/kg.	0.2 mg/kg at 12 hour intervals for three doses	27/32 (84.40%)	25/31 (80.60%)

From these studies, ibuprofen is as efficacious as indomethacin in the treatment of PDA.Additionally, it is significantly less likely to cause renal dysfunction indicated by infants treated with ibuprofen had higher creatinine clearance, urine volume and lower serum creatinine and BUN values than infants treated with indomethacin.

Recently, four randomised, controlled trials comparing indomethacin and ibuprofen for PDA closure were reported (87) as following.

Table 11 Ibuprofen versus indomethacin in the treatment of PDA.

Citation	Study group	Study type	Outcome	Key result	Comments
Lago et al (2002)	175 preterm neonates, GA≤34 wk, postnatal age 48-72 h with echo proven PDA. Very ill babies excluded. IBU (10 mg/kg iv [time 0], 5mg/kg iv [time 24,48 h]) v INDO [0.2 mg/kg iv q 12 h x 3)	RCT (level 1b) Jadad score: 2	Echo proven closure of PDA Oliguria Post-treatment serum creatinine	IBU = INDO for closure of PDA: ARR = 0.043 [95% CI -0.092 to 0.177] INDO more lokely to produce oliguria: p = 0.017, NNH = 7 INDO resulted in higher post-treatment creatinine (mean 89 μmol/l, SD 24) than IBU (mean 81 μmol/l, SD 20): p=0.03.	Only echocardiographers were noted to be blinded. Randomisation method not given. Allocation concealment by sealed envelope.
Van Overmeire et al (2000)	148 preterm neonates, GA≤32 wk, postnatal age 48-96 h with echo proven PDA. Very ill babies excluded. IBU (10 mg/kg iv [time 0], 5mg/kg iv [time 24,48 h]) v INDO [0.2 mg/kg iv q 12 h x 3)	RCT (level 1b) Jadad score: 2	Echo proven closure of PDA Oliguria Post-treatment serum creatinine Multiple logistic regression performed to determine predictors of oliguria	IBU = INDO for closure of PDA: ARR = 0.041 [95% CI -0.109 to 0.190]. INDO more lokely to produce oliguria: p = 0.03, NNH = 8 INDO resulted in higher post-treatment creatinine p=0.04. Independent predictors of oliguria were INDO treatment, high frequency ventilation, increased serum creatinine days 1-3, and lower ductal shunt velocity.	Only echocardiographers were noted to be blinded. Randomisation method not given. Allocation concealment by sealed opaque envelope
Van Overmeire et al (1997)	40 preterm neonates, GA≤33 wk, postnatal age 48-72 h with echo proven PDA. Very ill babies excluded. IBU (10 mg/kg iv [time 0], 5mg/kg iv [time 24,48 h]) v INDO [0.2 mg/kg iv q 12 h x 3)	RCT (level 1b) Jadad score: 2	Echo proven closure of PDA Oliguria Post-treatment serum creatinine	IBU = INDO for closure of PDA: ARR = 0.05 [95% CI -0.208 to 0.308] INDO more likely to produce oliguria: p = 0.02, NNH = 3 INDO = IBU p=0.07.	Randomisation method not given. Allocation concealment by sealed envelope. No blinding reported.
Supapanna-chart et al (2002)	18 preterm neonates (mean GA 30 wk with PDA based on clinical and X ray criteria. Very ill babies excluded. IBU (10 mg/kg po od x 3 days) v INDO [0.2 mg/kg po/iv q 12 h x 3 doses)	RCT (level 3b) Jadad score: 2	Clinical closure of PDA Oliguria Post-treatment serum creatinine	IBU = INDO for closure of PDA: ARR = 0.111 [95% CI -0.229 to 0.452] No significant difference after day 1. No significant difference.	Randomisation method not given. Allocation concealment by sealed envelope. No blinding reported. INDO group wasw mixed between babies receiving oral and intravenous treatment; no attempt at subset analysis.

All of these studies have no data about the methods of randomisation, the blinding of neonatologists, nurse, or pharmacists. All used cards in sealed envelops for allocation concealment. Each study clearly showed the equivalence of ibuprofen and indomethacin in the treatment of PDA. Additionally, three studies showed a significant increase in oliguria among patients treated with intravenous indomethacin, and two studies showed a significant increase in serum creatinine. No study showed difference in death, necrotizing enterocolitis, or progression of intracranial haemorrhage between the two groups. In conclusion, intravenous ibuprofen shows the same efficacy as intravenous indomethacin in PDA treatment in neonate, causes a smaller rise in serum creatinine and is less likely to develop oliguria (NNT = 6) than patients receiving intravenous indomethacin.

Considering major side effects on major organs, we can compare side effects between two drugs as followings.

1 Mesenteric blood flow

Pezzati et al. (29) evaluated the effect of ibuprofen and indomethacin on mesenteric and renal blood flow velocity in preterm infants.

Table 12. Comparison of indomethacin vs ibuprofen on the mesenteric and renal blood flow velocity at 30 and 120 min after treatment.

Drugs	**30 min after treatment**		**120 min after treatment**	
	Peak-systolic velocity Mean velocity End-diastolic velocity	**Relative vascular resistance**	**Peak-systolic velocity Mean velocity End-diastolic velocity**	**Relative vascular resistance**
Indomethacin	Significantly decreased	Significantly increased	Not returned to pretreatment value	Decreased
Ibuprofen	Not changed	Significantly increased but returned to pretreatment value at 60 min.	Significantly increased except PSV	

Ibuprofen didn't significantly reduce mesenteric blood flow and renal blood flow velocity.

Table 13. Comparison of indomethacin and ibuprofen on renal fuction.

Drugs	**Urine output**	**Serum creatinine**
Indomethacin	Significantly decreased on day 1 – day 3 but returned to pretreatment value on day 7	Significantly increased on day1 – day3
Ibuprofen	Significantly decreased after first dose	Didn't change significantly

The difference of urine output value between two groups could be observed on day2 and day3. Additionally, the difference of serum creatinine value could be seen on day 1 – day 3 and day 7.

2 Renal blood flow

In animal experiment by Chamaa et al. (34) showed that intravenous administration of ibuprofen (0.02-2 mg/kg body weight) to newborn rabbits caused a significant increase in renal vascular resistance and a fall in renal blood flow and glomerular filtration rate. In addition, a reduction in urine flow rate and urinary excretion of sodium could be found. Consequently, ibuprofen did not have significantly less renal side effects than indomethacin in animal. In contrast, many studies(27-29, 35) in preterm human reported less renal hemodynamic effect than indomethacin(27-29, 35).

3 Cerebral blood flow

Ibuprofen didn't affect cerebral hemodynamic (systolic peak velocity, end-diastolic velocity, mean flow velocity, resistance index, and pulsatility index remained constant in respect to pretreatment values) but it showed a significantly increase both in cerebral blood volume and cerebral blood flow velocity 60 minutes after administration(35). Additionally, ibuprofen didn't cause significant reduction of cerebral blood volume, oxidized cytochrome oxidase concentration, or cerebral blood flow velocity compared with indomethacin(36). Moreover, a significant increase of cerebral blood volume can be observed after 60 minutes in ibuprofen group (36)and

the mean arterial blood pressure, PaO_2 and SaO_2 were unchanged after ibuprofen was administered.

4 Bilirubin

According to the highly percentage bound to albumin, ibuprofen may compete with bilirubin in displacement from albumin. Cooper-Peel et al. (37) demonstrated that at clinically appropriate ibuprofen concentrations the free fraction of bilirubin is increased by a factor of 4. Thus ibuprofen may increase the risk of bilirubin encephalopathy when used in sick, premature infant. Ahlfors (82) also reported that ibuprofen have a great impact on bilirubin-albumin binding by increasing the free bilirubin level in pooled newborn plasma similar to sulfisoxazole(82). These reports suggested that the level of ibuprofen produced by IV form in PDA studies may high enough for causing this side effect.

5 Pulmonary hypertension

Gournay et al. (38) recently reported that ibuprofen administration in three premature newborn aged less than 28 weeks could cause respiratory distress syndrome and needed surfactant therapy. It is possible that ibuprofen given within the first 6 hours after birth could prevent the normal decrease in pulmonary vascular resistance because of cyclo-oxygenase inhibition. However, it might be resulted from a specific effect of ibuprofen. Consequently, neonatologists should aware and monitor this undesirable adverse effect.

Table 14.Advantages and disadvantages of ibuprofen therapy for patency of the ductus arteriosus.

Parameter	Advantages	Disadvantages
Therapeutic efficacy and availability	Appears to be as effective as indomethacin in closing PDA	No long term outcome studies No intravenous form universally commercially available
Effect on peripheral vascular beds	Probably less reduction in cetrebral blood flow than indomethacin Less impairment of renal and gastrointestinal haemodynamics than indomethacin	Reduction of prostaglandin E_2 production from cerebral microvessels, implying possible cerebral vasoconstriction
Bilirubin		Free bilirubin is increased by a factor of 4, possibly increasing the risk of bilirubin encephalopathy

Meta-analysis of randomized or quasi-randomized controlled trials (72) comparing ibuprofen to indomethacin for the treatment of PDA in preterm and/or low birth weight infants, eight studies including 509 patients were recruited. The characteristics of included studies could be summarized as followings.

Table 15 Summarization of the study comparing ibuprofen and indomethacin for PDA treatment.

	Akisu 2001 Italy	**Lago 2002 Italy**	**Mosca 1997 Italy**	**Patel 1995 UK**	**Patel 2000 UK**	**Plavka 2001 Czech**	**Van Overmeire 1997 Belgium**	**Van Overmeire 2000 Belgium**
Randomization								
Centre	Single	Single	Single	Single	Four	Three	Single	Single
Complete F/U	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Blinding of -Randomization -Intervention -Outcome	 Can't tell Can't tell Can't tell	 Can't tell No No	 Can't tell No	 Can't tell No No	 Yes Yes Yes	 Can't tell Can't tell Can't tell	 Yes No No	 Yes No No
Participants								
Ibuprofen group								
Number (girls+boys)	12 (5+7)	94 (42+52)	8 (4+4)	18	18 (9+9)	20	20	74
GA (wks)	32.1±1.2	28±2	29	26	26	27.6±2.3	29.0±2.4	29.0±2.3
BW (gm)	1706±187	1126±412	855		790	929±213	1270±450	1230±390
Surfactant use	7						15	56
Others	PDA diagnosis on day 3.9±0.5				Postnatal 8 days			
Indomethacin group								
Number	11	81	8	15	15	21	20	74
GA (wks)	31.9±1.3	29±3	28	26	26.7	26.9±1.7	28.7±1.9	29.0±2.1
BW (gm)	1645±190	1214±427	820		838	902±211	1210±360	1230±380
Surfactant use	7						19	63
Others	PDAdiagnosis on D 3.5±0.6				Postnatal 7 days			
Interventions								
Dose of IV ibuprofen (mg/kg)	10/5/5 q 24 h	10/5/5 q 24 h	10 single	5 single:12 infants 10 single:6 infants	10/5/5 q 24 h	8/8/8 q 24 h half dose q 24 h for 6 doses if PDA persisted	10/5/5 q 24 h	10/5/5 q 24 h
Dose of IVindomethacin (mg/kg)	0.2/0.2/0.2 q 12 h	0.2/0.2/0.2 q 12 h	0.2 single	0.1 single	0.2-0.25 q 12 h for 3 doses	0.2/0.2/0.2 q 24 h half dose q 24 h for 6 doses if PDA persisted	0.2/0.2/0.2 q 12 h	0.2/0.2/0.2 q 12 h
Measured Outcomes								
PDA closure	♣	♣	♣	♣	♣		♣	♣
PDA ligation							♣	♣
Ductalreopening						♣		
SCr	♣	♣				♣		
Oliguria		♣						♣
CBFV			♣		♣	♣		
CBV			♣	♣	♣			
COD					♣			
BP						♣		
Mortality	♣					♣	♣	♣ at D30
Sepsis	♣						♣	♣
NEC							♣	♣
GI bleed	♣							
ROP							♣	
IVH	♣							

The main outcomes reported in this systematic review were summarized:

Table.16 Summarization of the main outcomes of the study comparing ibuprofen and indomethacin for PDA treatment

Comparison or outcome	Studies	Participants	Statistical method	Effect size
Ibuprofen vs indomethacin				
Failure to close a PDA (after single or three doses)	7	509	RR (fixed), 95%CI RD (fixed), 95%CI	0.92 [0.69, 1.22] -0.02 [-0.10, 0.05]
All cause mortality	3	238	RR (fixed), 95%CI RD (fixed), 95%CI	0.96 [0.45, 2.04] 0.00 [-0.08, 0.07]
Neonatal mortality (during first 28/30 days of life)	1	148	RR (fixed), 95%CI RD (fixed), 95%CI	1.17 [0.41, 3.31] 0.021[-0.08, 0.10]
Infant mortality (death during the first year of life)	0	0	RR (fixed), 95%CI	No numeric data
Reopening of the ductus arteriosus	2	57	RR (fixed), 95%CI RD (fixed), 95%CI	1.74 [0.40, 7.46] 0.07 [-0.10, 0.24]
Need for surgical closure of the PDA	5	437	RR (fixed), 95%CI RD (fixed), 95%CI	1.03 [0.63, 1.68] 0.00 [-0.06, 0.07]
Need for treatment with indomethacin to close the PDA	0	0	RR (fixed), 95%CI	No numeric data
Duration of ventilator support	2	215	WMD (fixed), 95%CI	-1.26 [-5.01, 2.50]
Intraventricular haemorrhage (Grade I-IV)	2	63	RR (fixed), 95%CI RD (fixed), 95%CI	0.78 [0.23, 2.63] -0.03 [-0.21, 0.14]
Intraventricular haemorrhage (III-IV)	1	175	RR (fixed), 95%CI RD (fixed), 95%CI	1.23 [0.49, 3.09] 0.02 [-0.07, 0.11]
Periventricular leucomalacia (cystic)	2	323	RR (fixed), 95%CI RD (fixed), 95%CI	1.17 [0.52, 2.59] 0.01 [-0.04, 0.07]
Necrotizing enterocolitis (any stage)	3	363	RR (fixed), 95%CI RD (fixed), 95%CI	0.61 [0.24, 1.55] -0.02 [-0.07, 0.02
Decreased urine output (<1 cc/kg/hr)	2	323	RR (fixed), 95%CI RD (fixed), 95%CI NNT(fixed),95%CI	0.22 [0.09, 0.51] 0.13 [-0.20, -0.07] 9 [5-14]
Time to enteral feeds	2	323	WMD (fixed), 95%CI	1.74 [-1.21, 4.70]
Retinopathy of prematurity	1	40	RR (fixed), 95%CI RD (fixed), 95%CI	0.33 [0.01, 7.72] –0.05 [-0.18, 0.08]
Pneumothorax	0	0	RR (fixed), 95%CI	No numeric data
Chronic lung disease (at 28 days)	2	188	RR (fixed), 95%CI RD (fixed), 95%CI NNH(fixed), 95%CI	1.37 [1.01, 1.86] 0.14 [0.01, 0.29] 7 [3100]
Chronic lung disease (at 36 weeks corrected postnatal age)	1	175	RR (fixed), 95%CI RD (fixed), 95%CI	1.52 [0.83, 2.81] 0.08 [-0.03, 0.20]
Sepsis	4	386	RR (fixed), 95%CI RD (fixed), 95%CI	1.46 [0.72, 2.96] 0.03 [-0.02, 0.08]
Duration of hospitalization	1	175	WMD (fixed), 95%CI	-8.00[-18.59, 2.59]
Days on supplementary oxygen	2	215	WMD (fixed), 95%CI	6.84 [-3.12, 16.81]
Gastrointestinal bleed	1	23	RR (fixed), 95%CI RD (fixed), 95%CI	1.83 [0.19, 17.51] 0.08 [-0.20, 0.35]
Failure to close a PDA (after 3 doses)	6	476	RR (fixed), 95%CI RD (fixed), 95%CI	0.90 [0.66, 1.22] -0.03 [-0.11, 0.05]
Failure to close a PDA (after 3 doses, abstracts excluded)	5	435	RR (fixed), 95%CI RD (fixed), 95%CI	0.90 [0.66, 1.22] -0.03 [-0.11, 0.05]

1. There was no statistically significant difference between ibuprofen and indomethacin groups in failure to close a PDA after single or three doses.

2. There were no statistically significant difference in all cause mortality, neonatal mortality, ductal reopening, surgical ligation, duration of ventilator support, IVH, PVL, NEC, time to full enteral feeding, ROP, sepsis, duration of hospitalization or gastrointestinal bleeding.

3. The incidence of decreased urine output (< 1cc/kg/hr) was lower in the ibuprofen group than the indomethacin group. This was the only statistically significant clinical finding favouring ibuprofen.

4. Chronic lung disease defined as oxygen requirement at 28 days postnatally was statistically significantly more likely to occur in the ibuprofen group. Additionally, the similar trend at 36 weeks corrected gestational age was noted.

Because ibuprofen can cause CLD and PPHN, it does not appear to confer any benefits over indomethacin for the treatment of a PDA and indomethacin should remain the drug of choice for the treatment of a PDA. Although ibuprofen was likely to show higher incidence of chronic lung disease than indomethacin, the major renal side effect from ibuprofen was significantly less than indomethacin. Moreover, the efficacy in PDA closure was not different between both drugs. This meta-analysis was conducted in only eight studies and they might be a variation in the study design so the result should be carefully evaluated. The more number of randomized controlled trials is needed for the more accuracy in the interpretation of result.

Prophylactic ibuprofen

There are many studies (28, 39-42) to demonstrate its efficacy in achieving ductal closure when given prophylactically.

Table 17 Comparison of the study of PDA prophylaxis by ibuprofen.

Author/ Year	Gestational age	Intervention	Result	
			Efficacy	Adverse outcome
Varvarigou/ 1996	GA < 32 weeks	1. ibuprofen lysine 10 mg/kg IV followed by 5 mg/kg IV at 24 and 48 hours after first dose. 2. ibuprofen lysine 10 mg/kg IV single dose. 3. Placebo (saline)	- The incidence of PDA are 0% in three doses group, 54.5% in one dose group, and 63.6% in saline group. - Compared with the saline group, in three doses group had lower daily MAP, better oxygenation index, and required fewer day of ventilation.	- In three doses group, the incidence of IVH was not statistically significantly different from placebo. - The incidence of ROP, NEC, or mortality were not different among all groups.
Dani/ 2000	GA < 34weeks	1. Prophylactic group: 10 mg/kg IVwithin 24 hours ,followed by 5 mg/kg IV after 24 and 48 hours. 2. Rescue group: the same dosage after echocardiographical diagnosis of PDA.	- Prophylactic group developed a significant PDA lower than rescue group (8% vs 53%; P<0.0001) - 90% of patients in rescue group had a closure of ductus after ibuprofen treatment	-No difference in the incidence of BPD, IVH, ROP, and NEC between two groups. -No difference of mean serum creatinine level between the groups both before and after treatment.
De Carolis/ 2000	GA < 31weeks	1. Prophylaxis group: ibuprofen lysine IV 10 mg/kg followed by 5 mg/kg IV at 24 and 48 hours after first dose. 2. Control group: no intervention	Prophylaxis group had the rate of ductal closure at 72 hours higher than control group (87% vs 30.4%; P<0.0002) and less rate of back-up treatment than control group (13% vs 69.6%; P<0.0002)	-No significant differences in renal and gastrointestinal function, no haematological alterations, and no sepsis or NEC occurs. -Only food intolerance was more frequent (P=0.03) in the ibuprofen group in reapect to controls.
Hariprasad/ 2002	GA 28-35 weeks	All 13 neonates were received oral ibuprofen 10 mg/kg at first dose and 5 mg/kg at 12 hourly interval	11/13 patients'ductus closed after treatment	No significant side effects like oliguria or bleeding tendencies.
Raju/ 2000	GA 23-28 weeks	1. Study group: ibuprofen 10 mg/kg of loading dose followed by 5 mg/kg every 12 hours IV or oral depending on tolerance of enteral feed 2. Control group: no intervention	-Ventilator days in the study group is less than control group (34±25 vs 47±18; P=0.20) -Oxygen requirement, VEI were not significantly different between two groups.	BPD, NEC, and IVH were not statistical significantly different between two groups.

In the meta-analysis of randomized or quasi-randomized controlled trials comparing use of ibuprofen with placebo/no intervention or cyclooxygenase inhibitor drugs (indomethacin, mefenamic acid, etc) for the prevention of PDA in preterm

and/or low birth weight infants by Shah and Ohlsson (63), there were four trials that could be included in the following table.

Table 18 Comparison of RCTs for PDA prophylaxis in premature infants.

	Dani 2000 Italy	DeCarolis2000 Italy	Gournay2002 France	Van Overmeire2002 Belgium
Randomization				
Centre	Two	Single	Multicentre	Seven
Placebo	No	No		
Blinding of				
-Randomization	Yes	Can't tell	Can't tell	Unclear
-Intervention	No	No	Yes	Yes
-Outcome	No	Yes for 1^0outcome	Yes	Yes
Complete F/U	Yes	Yes	Can't tell	Yes
Participants				
Study period	Not stated	Apr,1996-Jul,1997	Mar,2001-Dec,2001	Not strated
Inclusion criteria	1. GA<34 wks 2. NCPAP with FiO_2> 30% or synchronized mechanical/high frequency ventilation	GA<31 wks	GA<28 wks Postnatal age<6 hr	GA<30 wks
Exclusion criteria	1. Bleeding disorder, Plt count<75000, SCr>1.5 2. Grade III or IV IVH		1. Congenital malformation 2. Shock or right to left ductal shunt 3. Cerebral complication 4. Bleeding disorder	
Enrolled within hours after birth	24	2		
Demographic data	Values presented as mean±SD	Values presented as mean±SD or as percentage	Not available	
Prophylaxis/Rescue				
N	40/40	25	65	172
GA (wks)	29.2±2.4 / 29.6±5.6	28.1±1.1		
BW (gm)	1231±445/1226±505	934±288		
Control				
N		25	66	186
GA (wks)		28.0±1.9		
BW (gm)		993±308		
Interventions				
Treatment group	-10/5/5 mg/kg q 24 h within first 24 h of life in prophylaxis group -10/5/5 mg/kg q 24 h after echocardiography in rescue group	10/5/5 mg/kg q 24 hr	10/5/5 mg/kg q 24 hr within 6 hr of life	10/5/5 mg/kg q 24 hr within 6 hr of life
Control group		No placebo/control treatment	Dose schedule as treatment group	Saline 1/0.5/0.5 ml/kg q 24 hr
Primary outcome(s)	Echocardiography on day 3, 7 and 21	PDA at 72 hr of age proven by echocardiography	Need for surgical ligation	IVH and PDA by echocardiography

The results of the review could be concluded as the following.

Table 19 Conclusion of randomized controlled trial (RCT) for PDA prophylaxis in premature infants

Comparison or outcome	Studies				No.	Statistical method	Effect size
	Dani 2000	DeCarolis 2000	Gournay 2002	Overmeire 2002			
1. Presence of PDA on third day of life (72 hrs of age)	√	√		√	488	RR (fixed), 95%CI RD (fixed), 95%CI NMT (fixed),95%CI	0.36 [0.26, 0.49] -0.29 [-0.37, -0.21] 3 [3, 5]
2. Presence of PDA on day 7	√				80	RR (fixed), 95%CI RD (fixed), 95%CI	0.20 [0.01, 4.04] -0.05 [-0.19, 0.03]
3. Mortality at 28 days	√	√			130	RR (fixed), 95%CI RD (fixed), 95%CI	1.0 [0.34, 2.98] 0.00 [-0.09, 0.09]
4. Mortality during hospital stay	√	√		√	130	RR (fixed), 95%CI RD (fixed), 95%CI	0.71 [0.15, 3.45] –0.02 [-0.09, 0.06]
5. CLD at 28 days		√			50	RR (fixed), 95%CI RD (fixed), 95%CI	0.83 [0.29, 2.38] –0.04 [-0.27, 0.19]
6. CLD at 36 wks CGA	√				80	RR (fixed), 95%CI RD (fixed), 95%CI	0.67 [0.12, 3.78] -0.03 [-0.13, 0.08]
7. Duration of mechanical ventilation (days)	√				80	WMD (fixed), 95%CI	0.40 [-6.57, 7.37]
8. Need for surgical ligation of PDA	√	√			130	RR (fixed), 95%CI RD (fixed), 95%CI	0.33 [0.04, 2.99] -0.03 [-0.10, 0.03]
9. Need for rescue medical treatment with COX inhibitors	√	√			130	RR (fixed), 95%CI RD (fixed), 95%CI NMT (fixed), 95%CI	0.10 [0.04, 0.28] -0.49 [-0.62, -0.36] 2 [2, 3]
10. IVH	√	√		√	488	RR (fixed), 95%CI RD (fixed), 95%CI	1.07 [0.59, 1.97] 0.01 [-0.04, 0.05]
11. PVL		√			50	RR (fixed), 95%CI RD (fixed), 95%CI	1.0 [0.15, 6.55] 0.00 [-0.15, 0.15]
12. NEC	√	√		√	130	RR (fixed), 95%CI RD (fixed), 95%CI	0.20 [0.01, 3.97] -0.03 [-0.09, 0.03]
13. GI hemorrhage	√				80	RR (fixed), 95%CI RD (fixed), 95%CI	3.00 [0.13, 71.51] 0.03 [-0.04, 0.09]
14. Time to reach full enteral feeds (days)	√				80	WMD (fixed), 95%CI	-0.30 [-4.47, 3.87]
15. Urine output after treatment (ml/kg/hr)	√				80	WMD (fixed), 95%CI	0.10 [-0.27, 0.47]
16. SCr (mg/dl)	√			√	438	WMD (fixed), 95%CI	0.11 [0.06, 0.17]
17. ROP	√				80	RR (fixed), 95%CI RD (fixed), 95%CI	0.77 [0.38, 1.55] -0.07 [-0.27, 0.12]
18. Sepsis		√			50	RR (fixed), 95%CI RD (fixed), 95%CI	1.0 [0.07, 15.12] 0.00 [-0.11, 0.11]
19. Pulmonary hypertension			√		131	RR (fixed), 95%CI RD (fixed), 95%CI	7.11 [0.37, 134.91] 0.05 [-0.01, 0.10]
20. Length of hospital stay	√				80	WMD (fixed), 95%CI	-3.50 [-15.55, 8.55]

1. The reduction of PDA incidence on day 3 is effective.

2. It did not find evidence of a statistically significant different in the mortality, need for surgical ligation, duration of hospitalization, CLD at 28 days or 36 weeks CGA,

duration of mechanical ventilation, IVH, PVL, NEC, GI hemorrhage, time to reach full enteral feed, ROP, urine output or sepsis between ibuprofen and placebo groups.
3. There was a statistically significant increase in the serum creatinine levels on day 3 of treatment in the prophylactic group as compared to the placebo group.
4. One trial reported on three infants in the ibuprofen group who developed pulmonary hypertension responsive to nitric oxide treatment. Because of this severe side effect, current evidence does not support the use of ibuprofen for prophylaxis of PDA. Additionally, the safety of prophylaxis needs to be established before it can replace indomethacin as a standard of practice.

Although, ibuprofen given intravenously to premature newborn infants is as effective as indomethacin in the treatment and prevention for PDA, the intravenous form of ibuprofen is not available in Thailand but the oral suspension is widely used in stead. There are many studies investigated the possibilities of oral ibuprofen for the treatment and prevention of PDA in Thai premature newborn infants as following.

Table 20 Comparison of the studies of PDA prophylaxis with ibuprofen in Thailand.

Author/ Study type/ Institute	Population / GA	Dose of ibuprofen	Dose of indomethacin	Efficacy result	Adverse outcome
Saowanee/Prospective randomized double blind controlled study/ Ramathibodi hospital	BW<1500g GA = 29wk	10 mg/kg (oral form) within the first 24 hours and then at 24, 48 hours after the first dose	-	-The incidence of symptomatic PDA in the placebo group(11/17) is higher than the treatment group(3/15) (66% vs 20%; RR=0.3, 95%CI=0.1-0.9) - The mean plasma concentration of ibuprofen 16.4 μg/ml at 2 hour after the first dose, 21.1 μg/ml before the second and the third dose, and 21.9 μg/ml 24 hour after the third dose.	BPD, IVH, NEC, and duration of ventilator support or complication were not significant difference between two group.
Anchalee/Prospective randomized parallel study/ Ramathibodi hospital	BW≤ 1750g GA< 34wks	10 mg/kg/dose(oral form) for three doses given at 24 hourly intervals	0.2mg/kg/dose (oral or intravenous form) for three doses given at 12 hourly intervals	-PDA was closed in 7/9(78%) infants given ibuprofen and 8/9(89%) infants given indomethacin (p>0.05) -The mean plasma concentration of ibuprofen was 28.05 μg/ml at 1 hour after the third dose	-Neonates in the ibuprofen group had more urine output than neonates in the indomethacin group. -The increment of BUN and creatinine, duration of ventilator support, BPD, IVH, NEC, and death were not significant in both groups.
Kesanat/ Prospective randomized clinical trial study/ QSNICH	BW< 1500g GA≤ 35wks	10 mg/kg/dose(oral form) for three doses given at 24 hourly intervals	0.1-0.25 mg/kg/dose based on body weight (intravenously) for three doses at 12 hourly interval	-The rate of ductal closure were 7/15(46.67%) in ibuprofen group and 10/15(66.67%) in indomethacin group (RR=0.669; 95%CI=0.328-1.364;p=0.462)	-The difference in using furosemide in indomethacin group was higher than than in ibuprofen group (11cases vs 3cases; p=0.009) -The incidence of NEC in indomethacin group was higher than in ibuprofen group but not statistically significant different (66.67% vs 40%)
Prasarn/ Prospective randomized parallel study/ Pramongkutklao hospital	BW< 1900g GA<37wks	10 mg/kg/dose(oral form) for three doses given at 24 hourly intervals	0.2mg/kg/dose (oral form) for three doses at 12 hourly interval	-5/6 (83.3%) in ibuprofen group and 5/5 (100%) in indomethacin group were diagnosed as RDS and the heart murmur sound disappeared 5/5 (100%) in ibuprofen group and 3/5 (60%) in indomethacin group. -Ibuprofen seem to show better outcome than indomethacin but not statistically significant because of small number of population.	-BUN and creatinine were higher than normal value one case in each group but returned to normal after withdrawing drugs. -IVH, NEC, and GI bleeding were not found in each group
Piyanuch/Maharajnakorn Chiangmai hospital	BW=1500g GA<31wks	10 mg/kg (oral form) within the first 24 hours followed with 5 mg/kg at 24, 48 hour after the first dose.	-	10/20 (50%) infants responsed to treatment proven by echocardiography. -7/20 (35%) infants could reduce ventilatory support. -3/20 (15%) infants did not respond to treatment.	-3/20 (15%) infants had transient oliguria. -2/20 (10%) infants showed the rising of serum creatinine (≥0.5 mg/dl) but returned to the normal value. -1/20 (5%) had GI bleeding. -1/20 (5%) had NEC after 2 wks treatment

These studies reported the same result as the foreign studies. Ibuprofen is as effective as indomethacin in closing PDA; however, it had indicated by less serum BUN and serum Cr and more urine output than indomethacin. The incidence of NEC, BPD, IVH, and duration of mechanical ventilator support were not significant different between group except in one study (45) which showed that the indomethacin group tended to have higher incidence of NEC than the ibuprofen group.

Recently, study by Heyman and colleagues (86) supported the effectiveness and the safety of oral ibuprofen suspension for PDA closure. The PDA closure rate was 95.5% (21/22 cases). Fourteen newborns were treated with 1 dose of ibuprofen, six were treated with 2 doses, and the remaining two were treated with 3 doses. No reopening of the ductus and no surgical ligation of the ductus were observed. Furthermore, there was no significant difference in serum creatinine level before and after treatment, no bronchopulmonary dysplasia, and no cases of bleeding tendency.

In conclusion, ibuprofen in form of the oral route was as efficacious and as safe as the intravenous route in PDA closure. However, the oral form affords several important advantages over the intravenous form. Firstly, intravenous ibuprofen is not available in many countries. Secondly, the required oral dose is of minimal volume. Next, oral administration is extremely simple. Finally, the oral form is cheaper than the intravenous one.

Safety of pediatric ibuprofen

There are many NSAIDs approved for use in children < 14 years of age but indications for which NSAIDs are regularly used in neonates include closure of the patent ductus arteriosus, prevention of IVH, reduction of fever and pain management.

At present, ibuprofen becomes more widely used in children such as antipyretic, anti-inflammatory, and treating of PDA. Many studies of safety and adverse drug reactions were reviewed. Lesco and colleagues (73) conducted the randomized double-blind acetaminophen-controlled trial in 84,192 children randomly assigned to receive 12 mg/kg of acetaminophen, 5 or 10 mg/kg of ibuprofen suspension. The risk of acute gastrointestinal bleeding was 7.2 per 100,000 (95%CI, 2 to 18 per 100,000) which was not statistically significantly different from

acetaminophen group (P=0.31). The observed risk for acute renal failure, anaphylaxis, and Reye's syndrome was zero per 100,000 each (95%CI, 0 to 5.4 per 100,000). No life threating or requirement of blood transfusion. However, low white blood cell count was significantly more common among ibuprofen-treated children (P=0.04). The low white blood cell counts were transient and generally mild (1.5 to 2.7 $\times 10^9$/L). The study concluded that with short-term use of ibuprofen in children, the risk of hospitalization for GI bleeding, renal failure or anaphylaxis was not increased. However, no data on the risk of severe outcome of prolonged ibuprofen use.

In children, the short-term use of ibuprofen has been shown to have no worse renal adverse event profile than acetaminophen. The prevalence of BUN level > 18 mg/dl and creatinine level > 0.7 mg/dl were slightly higher in the 108 children with a concomitant diagnosis of dehydration, both for ibuprofen and acetaminophen. However, the authors did not exclude the possibility that ibuprofen may cause acute renal failure in some children and provided no information on the risk of renal impairment in children after long-term use of ibuprofen or among children ineligible for the clinical trial such as those with severe dehydration (≥10% of body weight, preexisting chronic renal, endocrine, or neoplastic disease.

In the review article by Cuzzolin et al. (75), many non-steroidal anti-inflammatory agents can cause nephrotoxicity during prenatal, neonatal and children exposure. Causion should be taken when NSAIDs are administered to individuals with pre-existing renal problem or in association with other potentially nephrotoxic drugs.

Ibuprofen Pharmacokinetic in Premature Neonates and Children

The pharmacokinetic parameters of ibuprofen in different age group, clinical condition, dosage form, etc could be compared in the following table.

Table 21 Comparison of ibuprofen pharmacokinetic at different dosage form and age group.

	Aranda 1997	Konstan 1991	Brown 1992	Murry 1999	Kauffman 1992	Kelly 1992	Overmeire 2001	Sharma 2003	Capparelli 2002
Number			178	98			27	20	20
Age		6 – 12 yrs	3 mth – 12 yrs	12.5 yrs	3 mth – 10.4 yrs	11 mth-11.5 yrs	GA 28.6±1.9 wk	GA 30.45 wk	GA 27 wk
Weight				34.9 kg			1250±460g	1262.5	
Dosage form	IV	Tablet	Suspension	Tablet 88 Suspensio 7 Capsule 3	Liquid	Liquid	IV	Suspension	IV
Dose	10 mg/kg – day1 5 mg/kg – day2,3		5 and 10 mg/kg	24±1.9 mg/kg	6 mg/kg	6 mg/kg	10 mg/kg – day1 5 mg/kg – day2,3	10 mg/kg	10 mg/kg – day1 5 mg/kg – day2,3
Clinical status	Premature with PDA	CF vs Healthy	Febrile children	CFn			Premature with PDA	Premature newborn	Premature newborn
Cmax (mcg/ml)	D1 = 180.6±11.1 D2 = 116.6±54.5 D3 = 113.6±58.2	48±17 vs 66±10	5 mg/kg=19.03 10 mg/kg=34.35	83±22	35.8±16.7	26.67	D3=43.5 D5=42.4	20.09	
Tmax		66±20 vs 60±24 min	5 mg/kg=1.60 h 10 mg/kg=1.54 h	1.77±0.7 hrs	0.7±0.5 hr	54.05 min		3.0 hr	
AUC		6.1±1.7 vs 11.3±3.4 mg.min/ml	5 mg/kg=71.12 µg.h/ml 10 mg/kg=115.76 µg.h/ml		102.6±35.2 mg.hr/l		D3=524 mg.hr/l D5=447 mg.hr/l	402.60 µg.h/ml	
CL/F	Cl 2.06±0.33 ml/kg/hr	2.3±0.6 vs 1.3 ± 0.2 ml/min.kg^{-1}	5 mg/kg=0.089 l/kg/hr 10 mg/kg=0.099 l/kg/hr	45.5±14.7 ml/min/m^2		Cl 0.96 ml/min/kg	CrCl D3=14.0 ml/min D5=12.4 ml/min		Cl 1.46ml/hr/kg
T1/2	30.5±4.2 hrs	92±27 vs 86±17min	5 mg/kg=1.65 h 10 mg/kg=1.48 h		α=0.3±0.3hr β=1.6±0.7hr	α=16.3min β=118.2min	D3=43.1 hr D5=26.8 hr	15.72 hr	30 hr
Vd/F	62.1±3.9 ml/kg	291±91 vs 158±43 ml/kg	5 mg/kg=0.182 l/kg 10 mg/kg=0.217 l/kg	5.3±1.2 l/m^2		164 ml/kg	Vd_c D3=0.244 l/kg D5=0.171 l/kg		Vd_{ss}=63 ml/kg
Ke	0.032±0.004 hr^{-1}		5 mg/kg=0.54 hr^{-1} 10 mg/kg=0.55 hr^{-1}	0.51±0.12 hr^{-1}	0.564±0.688 hr^{-1}				
Ka			5 mg/kg=8.42 hr^{-1} 10 mg/kg=6.30 hr^{-1}	1.91±0.98 hr		0.043 min^{-1}			

The pharmacokinetic study of intravenously administered ibuprofen in premature infants with patent ductus arteriosus by Van Overmeire et al. (54) showed that there was a change in some pharmacokinetic parameters between the first and the third dose (day3 and day5). The volume of distribution of the central compartment (Vd_c) was significantly decreased (0.244 vs 0.171 l/kg; P=0.03) and area under the plasma concentration-time curve (524 vs 447 mg.h/l; P=0.01). However, total body clearance, plasma half-life, and peak plasma concentration did not change significantly. Like gentamicin and indomethacin, the decrease in volume of distribution may be due to fluid overload in the presence of patent ductus arteriosus(66). There was a wide interpatient variability in ibuprofen pharmacokinetics in premature infants with respiratory distress syndrome and patent ductus arteriosus, a significant changes may occur between day3 and day5 after birth in those infants with a closing ductus.

In addition, Aranda et al. (55) reported the pharmacokinetic profile of intravenous ibuprofen lysine showed that the elimination is slower than older children and adults because the plasma half-life is prolonged more than 10-fold compared to the older babies. In addition, the percentage bound ibuprofen was significantly lower in full term cord plasma (94.98 ± 0.39%, n=26) compared to adult plasma protein (mean ± SE = 98.73 ± 0.31%, n=8, P<0.0001). This could be inferred that more free or unbound drug was present in the newborn resulting in a more intense pharmacologic effect relative to adults. Lastly, ibuprofen suspension pharmacokinetics was reported by Sharma et al(81). A single dose of 10 mg/kg ibuprofen suspension in 20 premature neonates produced Cmax 20.09±3.33 μg/ml, Tmax 3.0±0.22 hrs, T1/2 15.72±3.76 hrs, and $AUC_{0-\infty}$ 402.60±79.67 μg.hr/ml. These parameters showed an obvious large interpatient variability. Some variables had been reported to influence on the pharmacokinetic disposition of ibuprofen. $AUC_{0-\infty}$ and plasma T1/2 were in the higher range for male infants, as well as infants of GA less than or equal to 30 wks and infant with birth weight less than or equal to 1200 g, but the difference was not reach the statistically significant.

From the above three studies, pharmacokinetic parameters in premature infants showed a large interpatient variability. The absorption and elimination processes were

obviously slower than younger children and adult. The immaturity of drug biodisposition organs was suggested to explain these differences.

Firstly, the pharmacokinetic condition of ibuprofen in older children have also been studied. Konstan and colleagues (56) investigated pharmacokinetic of ibuprofen tablet in children 6-12 years with cystic fibrosis. Compared with values in healthy children who received a similar dose, mean plasma total ibuprofen concentrations were lower in children with cystic fibrosis because of an increase in apparent clearance and larger apparent V_{area}. Time to reach C_{max} and $T_{1/2}$ were similar in both groups. All of these parameters are dose independent.

Kauffman et al. (57) studied ibuprofen pharmacokinetics and antipyretic effect in infants and children aged 3 months to 10.4 years. Mean pharmacokinetic values of single dose 6 mg/kg liquid ibuprofen could be stated that ibuprofen was rapidly absorbed from the gastrointestinal tract after administration. The age of the child did not significantly influence the rate of absorption of ibuprofen but there was a significant age effect on antipyretic response and time to onset of antipyresis. The mean time to onset of antipyresis and the mean maximum decrease in temperature in the youngest children were faster and higher than the oldest children respectively.

Kelly et al. (58) studied the pharmacokinetics of racemic ibuprofen and its stereoisomers in children age ranged from 11 months to 11.5 years.The clearance of the R(-) isomer was greater than the total ibuprofen, which would be expected given the additional elimination pathway of 60% bioconversion of the R(-) to the S(+) isomer. The shorter elimination half-life of 88.2 minutes for R(-) vs 118.2 minutes for total ibuprofen also supported the previous report. The clearance of other isomer, S(+), was based on the assumption that the administered dose was4800 μg/kg; 50% of the 6000 μg/kg racemic dose (3000μg/kg) plus 60% conversion (1800μg/kg of R(-) to S(+). The nonconversion clearance of S(+) must be greater than that of R(-) because of the equality clearance values of R(-) and S(+). The maximum concentration of each isomer almost equal and maximum concentration of each isomer was about half of total ibuprofen. From these comparative studies (54-58, 80-81), we can conclude the difference in pharmacokinetic

properties according to the age and clinical status as following. Firstly, the elimination half-life in younger children is slower than older children and adults as reflected by

prolonged plasma half-life about 10-fold to 20-fold compared to the older babies. This could be inferred that more free drug was present in the newborn resulting in a more intense pharmacologic effect such as faster mean time to onset of antipyresis and higher mean maximum decrease in temperature (57) relative to older children and adults. Next, the immaturational activity of several hepatic metabolizing enzymes, the functional polymorphism of gene, and the variability in absorption observed in younger children especially premature infants could be the causes of these observations that supported by Brown's study (84). His study confirmed an effect of age on pharmacokinetic parameters. For both ibuprofen 5 and 10 mg/kg, the $AUC_{0\rightarrow\infty}$ was larger and Vd/F and Cl_p/F were less in the older than in the younger age group because age-dependent protein binding may be operative as suggested by the authors. Finally, mean plasma concentration in children with cystic fibrosis was lower than healthy children who received a similar dose because volume of distribution and clearance were increased. Individualized dosages and therapeutic drug monitoring may be required to ensure plasma concentrations considered necessary to prevent pulmonary deterioration in patients with cystic fibrosis (56-80).

In conclusion, pharmacokinetics parameters in neonates could be affected by many factors. The major factor is immaturity of drug biodisposition organs in premature neonates and the difference in many physiologic factors in different ages of life. The disease state such as cystic fibrosis could also affect the pharmacokinetics parameters. Therapeutic drug monitoring especially for narrow therapeutic drug may be necessary for premature infants in order to reach a maximum pharmacologic action with less adverse effect of the drug.

Clinical Pharmacokinetic properties of ibuprofen

Ibuprofen, (±)-(R,S)-2-(4-isobutylphenyl)-proprionic acid, is a chiral 2-arylproprionic acid (2-APA) derivative nonsteroidal anti-inflammatory drug. It is a potent inhibitor of prostaglandin synthesis with the S-(+)-enantiomer possessing the majority of pharmacological activity that is about 160 times more potent than R-(-)-ibuprofen in inhibiting prostaglandin synthesis.

Absorption

Ibuprofen lysine salt produces peak plasma drug concentrations (C_{max}) significantly earlier and higher than ibuprofen acid or granule because of the faster dissolution of ibuprofen lysinate. It has an absolute bioavailability approaching 100%.

The solution forms are absorbed more quickly (t_{max} < 0.25 hours) than the tablet (t_{max} ≈ 2 hours). However, the rate and extent of absorption from suspensions are significantly less than that of tablets and they are less bioavailable than the solution.

When administered with food, the t_{max} was about 20% lower and was attained more slowly (after 1.5 to 3 hours). In addition, high sugar loads causing a delay in gastric emptying were postulated to contribute in the delay in ibuprofen absorption. For example, a Coca-Cola® solution could reduce t_{max} , C_{max} , and AUC.

AUC versus dose plots from the ibuprofen 250 to 1,200 mg dosage range demonstrate a nonlinear relationship between total drug concentrations with dose and a more linear relationship between free concentrations. This non-linearity for total concentrations is characterized by a smaller than expected increase in the AUC for the total drug dose. It is possibly due to a saturation of protein binding rather than impaired absorption, as urinary recovery data appeared to be dose-independent.

Distribution

The apparent volume of distribution (V_d / F) after oral administration was between 0.1 to 0.2 l/kg in humans which approximates plasma volume and suggests tissue binding is appreciably less than plasma protein binding. The V_d / F based on total concentrations increases significantly with dose; however, there is a lack of dose-related changes in free drug V_d / F.

Ibuprofen is extensively more than 98% bound to whole human plasma and purified albumin at therapeutic concentrations. An apparent linear dose-response relationship exists between the amount of drug administered and the area under the serum concentration time curve after single doses of 200-800 mg. At low dose (400mg) there were no significant differences between oral and intravenous serum protein binding for the enantiomers of ibuprofen. Because of the saturation of plasma protein binding, there is an apparent decrease in AUC/dose values after higher doses, which contributes to nonlinear kinetics of ibuprofen. In a dose ranging 50-1,200 mg,

the AUC versus dose relationship were nonlinear at higher doses (above 800mg). Considering each enantiomer relationship between drug dose and AUC, a linear relationship was obtained for S-(+)-ibuprofen while a curvilinear relationship was observed for R-(-)enantiomer. The protein binding of each enantiomer was stereoselective and mutually competitive, as well as nonlinear; however, no difference was observed in the binding capacity as a function of chirality. The intrinsic binding of R-(-)-ibuprofen was greater than that of S-(+)-ibuprofen and the unbound fraction was greater for S-(+)-ibuprofen versus R-(+)-ibuprofen after a given dose of R-(-)-ibuprofen or racemate.

Metabolism

Ibuprofen is extensively metabolized via formation of the major metabolites 2-[4-(2-hydroxy-2-methylpropyl) phenyl] proprionic acid (2-hydroxyibuprofen) and 2-[3-(2-carboxypropyl)phenyl] proprionic acid (carboxy ibuprofen). 1-Hydroxy ibuprofen and 3-hydroxy ibuprofen have been found in urine in very small concentrations. Additionally, the hydroxy and carboxy metabolites have no apparent pharmacological activity.

Cytochrome P450 (CYP) 2C9 has been identified as the most important catalyst for formation of all the oxidative metabolites of R-(-) and S-(+)-ibuprofen. CYP2C9 is the major CYP mediating the 2 and 3-hydroxylations of R- and S-ibuprofen in the liver of Caucasians. CYP2C8 may also play a role in these biotransformations.

A major metabolic pathway of ibuprofen is conjugation with glucuronic acid to yield acylglucuronides. Additionally, covalent binding of the ibuprofen glucuronide to plasma protein may be an important cause of toxicity and anaphylactic reactions.

Following oral administration of ibuprofen, about 80% of the dose is recovered in urine as the hydroxy and carboxy metabolites, respectively, as a mixture of conjugated and unconjugated forms.

Elimination

Excretion in the urine

The excretion of drug and metabolites occurs rapididly in both urine and faeces. The urinary excretion of the metabolites showed that R-(-)-ibuprofen is inverted to S-(+)-ibuprofen with 80% of the urinary products as S-(+)-ibuprofen and

20% as R-(-)-ibuprofen and 54% as S-(+)-hydroxy ibuprofen and 46% as R-(-)-hydroxy ibuprofen. After administration of the racemate 71% is S-(+)-ibuprofen, 30% is R-(-)-ibuprofen, 71% is S-(+)-hydroxy ibuprofen and 29% is R-(-)-hydroxy ibuprofen. Total recovery in urine of ibuprofen and its metabolites is between 70-90% of the administered dose after a 24-hour collection and it is not significantly different between intravenous (≈81%) and oral administration (≈87%).

Urinary recoveries of hydroxy and carboxy ibuprofen between doses of 100-1200 mg showed no significant change with dose. Total recovery is about 80%, which is 45% carboxy ibuprofen, 25% hydroxy ibuprofen and about 12% ibuprofen. Following administration of R-(-)-ibuprofen there is a total recovery of hydroxy ibuprofen (21.7%) and carboxy ibuprofen (33.4%); however, when the dose of S-(+)-ibuprofen is considered, the total recovery is 28.3 and 43.3% respectively. Consequently, the recovery of carboxy metabolite is significantly greater for S-(+)-ibuprofen than the others implying that significant substrate stereoselectively exists for carboxy ibuprofen formation. In other words, we can state that the total urinary recovery of ibuprofen and its metabolites after S-(+)-ibuprofen (82%) is significantly greater than after R-(-)-ibuprofen (66%) and racemic doses (67%). Moreover, the urinary recovery of carboxy ibuprofen is significantly greater for S-(+)-ibuprofen relative to the R-(-)-enantiomer or racemate.

Clearance

The $t_{1/2\ \beta}$ and clearance values of the enantiomers administered separately are comparable to the racemate. The mean residence time of S-(+)-ibuprofen after administrations of R-(-) and racemic ibuprofen is significantly longer than after administration of the pure S-(+)-enantiomer. Considering each enantiomer, clearance of S-(+)-ibuprofen is greater than R-(-)-ibuprofen and likewise the unbound partial clearances of S-(+)-ibuprofen is greater.

Pharmacokinetic model of ibuprofen

The pharmacokinetic model for ibuprofen is elucidated from concentration-time data obtained by 15 normal volunteers (49). Ibuprofen is best describes a two-compartment open model, with first-order absorption as figure 4.

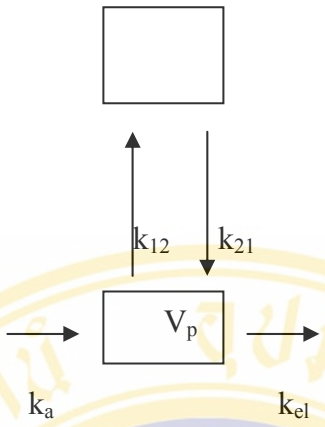

Figure 4 A two-compartment open model for ibuprofen pharmacokinetics based on individual subject concentration-time data. k_{12} ,k_{21} = intercompartmental rate constants; k_a = absorption rate constant; k_{el} = elimination rate constant. The half-life was 2.04 hours and the distribution volume of compartment one was 6.35 liters.

This pharmacokinetic model has been applied to total plasma concentrations after the administration of 400, 800, and 1200 mg tablet to young volunteers (49-50). On the basis of this analysis, absorption of ibuprofen tablets is not a simple first-order process. Absorption profiles after ingestion of one tablet were S-shape, whereas those after two or three tablets had partial linear segments, indicating zero-order absorption According to this model, the volume of distribution was 6.25 liters. This small value suggested that ibuprofen is highly bound to plasma protein and not extensively distributed.

Dose-related pharmacokinetics

Lockwood et al. (50, 52) study the ibuprofen pharmacokinetics of 15 volunteers. Each subject was assigned to one of three groups, and received one of four treatments (A through D) sequentially over a four weeks period. Treatment A, B, and C consisted of one, two, three 400mg ibuprofen tablet, respectively. In the final week of the study, all subjects received treatment D, which was 20 ml of an oral solution of ibuprofen (20mg/ml). In all subjects, there was a nonlinear relation between the administered dose and the area under the total ibuprofen plasma concentration time curve.

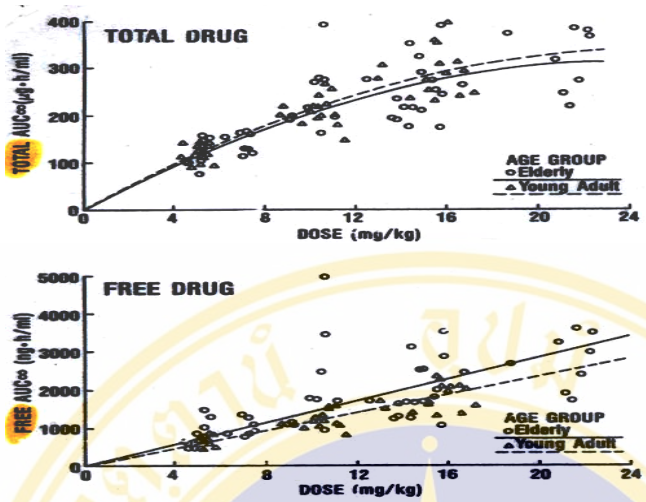

Figure 5 Plots of total and free ibuprofen in the area under the time-concentration curve (AUC_∞) versus dose (mg/kg) in 17 elderly and 15 young adult subjects. The curves drawn through the plots are least-squares parabolas (total drug) of lines (free drug) forced through the origin.

The AUC versus dose plots shown in the above figure emphasize the nonlinear relation between total drug levels with dose and the more linear relation for free concentration. These results are consistent with those reported in young adult subjects.

Total urinary recovery of ibuprofen as unchanged drug and its major metabolites was about 80 percent (range 76 to 85 percent) and consistent from treatment to treatment. The urinary excretion of ibuprofen and its metabolites is a linear function of dose, and any dose dependency associated with the absorption process is minor. Therefore, the disproportionate increase in total plasma levels may be due to nonlinear protein binding of ibuprofen as shown in following table. Mean ratios decrease significantly with increasing dose, indicating that the binding of ibuprofen is nonlinear.

Table 22 Bound /Free ratios (C_b / C_f) of ibuprofen as a function of dose

Parameter	**Solution (n = 100)**	**One 400-mg tablet (n = 102)**	**Two 400-mg tablets (n = 100)**	**Three 400-mg tablets (n = 104)**
Mean	183	196	173	166
Range	134-233	127-376	116-232	112.9-293.0
SD	22.6	39.4	27.4	28.8
CV(%)	12.3	20.1	15.8	17.4

Drug interactions

Aspirin. According to unpublished data, the multiple-dose administration of 1,300 mg/day of aspirin concomitantly with 2,400 mg/day of ibuprofen for five days reduced ibuprofen plasma levels to less than half those observed with ibuprofen alone as following figure 6.

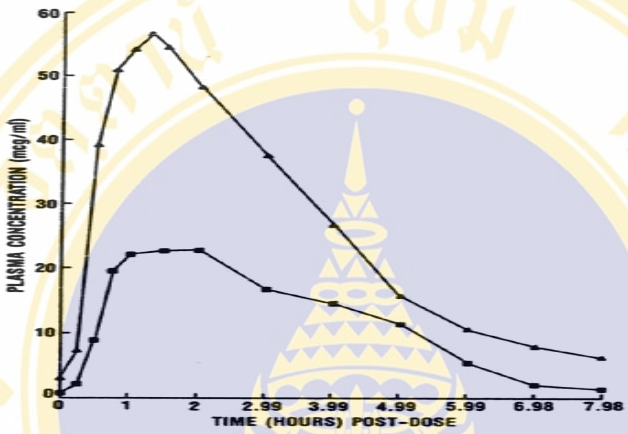

Figure 6. Effect of aspirin on ibuprofen plasma levels. Black squares = ibuprofen + aspirin; black triangles = ibuprofen alone.

Although the clinical significance of the ibuprofen/aspirin interaction has not been proved, the potential for an alteration in the efficacy of these drugs does exist.

Acetaminophen. The bioavailability and pharmacokinetics of ibuprofen are unaltered when given in combination with multiple doses of acetaminophen. When 400 mg of ibuprofen and 650 mg of acetaminophen given together every six hours for eight dose; after dose eight, serial samples were collected for 12 hours. The steady-state blood levels of acetaminophen and ibuprofen were virtually superimposable when these drugs were administered together. Because of no drug interaction, acetaminophen can be considered an appropriate over-the-counter analgesic in patients taking ibuprofen. The pharmacokinetics of ibuprofen can accurately be evaluated, even when acetaminophen has been administered as a rescue measure.

Pharmacokinetics of ingested xenobiotics in neonates: A comparison with adults

The main concern of pharmacokinetics is drug disposition. The four principles of drug disposition include absorption, distribution, metabolism, and excretion. The following information applies to the general pediatric patient. Information pertaining to neonates in the following sections applies to the term neonates. Preterm neonates should be treated as a special patient population.

Drug absorption

The gastrointestinal absorption of drugs is influenced by numerous factors. With oral administration, drug absorption depends upon both the physiochemical properties of the drug and a variety of patient factors that can be listed in table 23.

Table 23. Factors affecting drug absorption

Physicochemical factors
* Disintegration of tablets or solid phase and release characteristics for sustained-release formulation.
* Dissolution of the drug in gastric or intestinal fluid.
* pK_a and number of ionisable groups.
* Degree of lipid solubility of the lipid-soluble form.
* Molecular weight.
Patient factors
* Surface area available for absorption.
* Gastric and duodenal pH.
* Gastric emptying time.
* Bile salt pool size.
* Bacterial colonization of the gastrointestinal tract.
* underlying disease states.

However, the gastric emptying time and the gastric pH are very important for drug whose absorption is controlled a pH-dependent passive diffusion process.

Gastric pH

Gastric pH is usually between 6 and 8 (neutral) at birth due to the presence of amniotic fluid in the stomach, falls rapidly to 1.5 to 3.0 within a few hours to neutral in the following 24 hours (59-62, 76-77). After the first 24 hours, there is a state of relative achlorhydria, which may persist for 10 to 15 days (59-62, 76-77). However, acid is rarely present and the initial decrease in pH do not occur in premature neonates because of immaturity of the secretory mechanisms and immaturity of the gastric

mucosa, which may persist for 10-15 days. Consequently, the state of relative achlorhydria occurs more often in the preterm than term neonate. The highest hydrochloric acid concentration in premature infants with birthweights ranging from 1 to 2.1 kg was observed on day 4 and there were no relationship between birthweight and the amount of acid secreted by the stomach or the degree of development of the gastric mucosa.

Gastric emptying time (GET)

GET is delayed in the immediate newborn period 10 to 24 hours, for both full term and preterm neonates (59-62, 76-77). It reaches adult values only at 6 to 8 month of age. The rate of GET is variable during the neonatal period and is affected by numerous factors such as gestational maturity, postnatal age, type of feeding and clinical disease state.

Table 24. Factors affecting gastric emptying rate.

Increase	Decrease	No effect
Human milk	Prematurity	Osmolality
Hypocaloric feedings	Gastro-esophageal reflux Respiratory distess syndrome Congenital heart disease Long chain fatty acids	Posture

With human milk feedings, there is a characteristic biphasic pattern of gastric emptying in both preterm and term infants, with an initial rapid phase followed by a slower, prolonged second phase (60). GET is also affected by the composition of the meal. In premature neonates, GET is slower with increasing caloric density. For example, infants given a 10% glucose solution have slower GET compared with a 5% glucose solution (59-60). Slower emptying is seen in feedings with long-chain fatty acids than with medium-chain triglycerides (59-60). The disposition of drugs that undergo enterohepatic recirculation after conjugation with glucuronic acid may be affected because flow intraluminal bile acid concentration. This is related to a reduced bile acid pool in newborns (50% of adult values) and premature neonates (33% to 50% of values for full-term neonates), which is caused by ineffective ileal reabsorption of

bile acids and increased jejunal permeability to bile acids. The osmolality of the meal does not affect gastric emptying rate; for example, premature infants fed isocaloric food formulas do not show a difference in gastric emptying rates (59-62, 76-77). In addition, changes in body position during or after feeding do not appear to affect gastric emptying.

Fluid secreted by exocrine pacreas (59-62, 76-77)

The amount of fluids secreted into the intestine increases with maturation. In the preterm neonate 34-36 weeks, lipase activity was only half that in term neonates but a further 10-fold increase of lipase activity could be found in infant aged between birth and nine months of age. Amylase activity is very low at birth and increases a 200-fold by the age of 9 months resulting in a less adequate utilization of starches by young infants compared to older infants and children.

Bile secretion

In preterm neonates, bile flow is low compared to adult levels and the bile is composed of only primary bile acids (cholic acid and chenodeoxycholic acid) preferably conjugated to taurine.

Table25 Biliary bile salt composition in neonates, infants and adults.

Age	Ratio of glycine : taurine	Choline : Chenodeoxycholine : Deoxycholine
Neonate		
1-4 days	0.47	2.5:1:0
5-7 days	0.95	2.5:1:0
Infant		
7-12 months	2.4	1.1:1:0
Adult	3.1	1.2:1:0.6

The cholic acid and chenodeoxycholic acid ratio was high at birth and decreased with increasing postnatal age. During the first weeks of life, the bile acids were

preferentially conjugated with taurine but the taurine/glycine ratio decreased also with postnatal age.

Small intestine

In neonates, the rate of cell maturation from crypt to villus type was approximately 2-fold lower compared to adults (59-63, 76-77. The small intestinal function further matures throughout prenatal and postnatal life. In children the villi of the small intestine tend to be broad leaf-shaped rather than finger-shaped projections as in adults, implying a relative smaller functional surface area of the small intestine in neonates (76-77).

There is an important difference between neonates and older individual in the type and degree of bacterial colonization of the gut. At birth, the intestine is virtually sterile and a rapid colonization occurs with a flora that is different in breast-fed and formula-fed infants. A further change in bacterial flora occurs at the time of weaning (4-6 months) which is important for the hydrolysis of compounds that are conjugated and secreted in the bile so that unconjugated compound can be absorbed by the intestinal epithelium (e.g. conjugated bile acids). On the other hand, the neonatal gut is capable of converting glucuronides, excreted into the gastrointestinal tract from bile, to their unconjugated and hence enterohepatic reabsorbable form by the presence of β-glucuronidase.

Transporters

Active drug transport across cellular membranes is an important process involved in absorption, disposition and excretion of compounds. It is essentially important in liver, gut, kidney and the blood brain barrier.

Inorganic sulfate (Si) is an important anion that involved in many physiological and pharmacological processes, including activation and detoxification of many endogenous and exogenous compounds (76). The steady-state serum Si levels in humans vary during development. Neonates and infants had an elevated mean serum Si concentration compared with adult levels. The higher serum Si levels in the neonates may in part be attributed to a difference in amino acid and protein intake and the fact that the glomerular filtration rate is lower in young infants than adults. It is expected that neonates and infants have a reduced number of Si transporters (76). Since the total renal tubular mass and brush border and basolateral surface areas of the

proximal tubule are much smaller at birth than in adulthood and the proximal tubular segment where the majority of Si reabsorption occurs is poorly developed in neonates and infants compared to the adult.

A group of transporters that are much more important for the transcellular transport of exogenous molecules are the multidrug resistance protein (P-glycoprotein) and related transporters (76-77). These are members of the ATP binding cassette (ABC) superfamily of transport proteins which is the largest and most widespread protein superfamily known. Human P-glycoprotein has been detected in the apical surface of epithelial cells from excretory organs, such as the bile canalicular membrane of hepatocytes in the liver, the proximal tubules in the kidney and in enterocytes lining the wall of the intestines. The presence of P-glycoprotein in the intestine is an important factor in mediating direct efflux through the intestinal wall and/or limits the re-uptake after hepatobiliary excretion. Additionally, it can reduce the absorption of compounds following oral administration, thus protecting the host against orally ingested toxins.

In human fetal tissue, the expression of mdrl-mRNA could already be demonstrated in the embryonic phase of human development, after 7 weeks of gestation (76-77). However, there are some differences in tissue distribution between fetal and adult. The expression of P-glycoprotein in intestine was limited at birth and increased significantly with maturation in intestine. In contrast, hepatic and renal P-glycoprotein expression was at adult levels at birth. Because intestinal P-glycoprotein expression is not at adult levels at birth, the protection against orally ingested toxins might be less than in adults (76-77). It may speculate that the expression is only induced after challenges by toxins that are a substrate for P-glycoprotein so the expression at birth is low.

The other important transporter is the organic anion transporter (OAT) family. It plays a critical role in protecting against the toxic effects of anionic substances such as antibiotics, NSAIDs, and 2, 4-dichlorophenoxyacetic acid toxin by removing such substances from the blood via a transport mechanism found in the basolateral membrane of renal epithelial cells (76-77). The OA secretion is low at birth and increases over the first few weeks of neonatal life and the declines to adult levels. The

increase in secretion was disproportionate to the increase in renal mass and was thought to reflect the specific maturation of the organic anion transport system.

By conclusion, we can summarize age-dependent differences that impact drug absorption from extravascular routes of administration (77) in the table 26.

Table 26 Impact of physiologic change to pharmacokinetic parameter in three different age groups.

	Neonate	**Infants**	**Children**
Physiologic aleration			
Gastric emptying time	Irregular	Increased	Slightly increased
Gastric pH	>5	4-2	Normal (2-3)
Intestinal motility	Reduced	Increased	Slightly increased
Intestinal surface area	Reduced	Near adult	Adult pattern
Biliary function	Immature	Near adult	Adult pattern
Muscular blood flow	Reduced	Increased	Adult pattern
Skin permeability	Increased	Increased	Near adult pattern
Possible pharmacokinetic consequences			
Oral absorption	Erratic-reduced	↑Rate*	Near adult pattern
Intramuscular absorption	Variable	Increased	Adult pattern
Percutaneous absorption	Increased	Increased	Near adult pattern
Rectal absorption	Very efficient	Efficient	Near adult pattern
Presystemic clearance	<Adult	>Adult	>Adult (↑rate*)

* The direction of alteration is given relative to the expected normal adult pattern

Drug protein binding and drug distribution.

There are many factors that affect the rate and the amount of drug distribution. For example, the physicochemical properties of the drug, the local pH, relative regional blood flow, the percentage of extracellular water, the adipose tissue content and the degree of binding to plasma and tissue proteins. The important factors can be described as following.

Vascular perfusion. Persistant fetal circulation, which can occur in neonates with a number of conditions such as respiratory distress and postasphyxial syndromes can markedly divert blood from the lungs to other tissues and organs because of a right-to-left vascular shunt (59-62, 76).

Body composition. In both premature and full term neonates, the organs have a relative and absolute size which is different from those of older children and adults. For example, the liver is much larger in neonates in relation to their body weight (59-62). Neonates have a relatively underdeveloped muscular system and a high proportion of the body weight is formed by head (59-62). In the foetus and neonate, the blood brain barrier is underdeveloped, the myelin content of the brain is lower and the cerebral blood flow is relatively larger than in adults (59-62, 77). Therefore, higher exposure of the brain to small hydrophilic xenobiotics is expected in, especially, neonates (76).

The total body water is greater with a predominance of extracellular over intracellular water; the skeletal muscle mass represents only 20 to 25% of total bodyweight (59-62).

Table 27. Effect of age on organ weight as percentage of body weight

Organ	Organ weight as percent of body weight		
	Fetus	Full-term newborn	Adult
Skeletal muscle	25.0	25.0	40.0
Skin	13.0	4.0	6.0
Heart	0.6	0.5	0.4
Liver	4.0	5.0	2.0
Kidneys	0.7	1.0	0.5
Brain	13.0	12.0	2.0
Other (e.g.bone)	43.7	52.5	49.1

Table 28. Fluid compartment size as a function of age (as a percentage of bodyweight).

Age	Total body water	Extracellular fluid	Intracellular fluid
Less than 3-month fetus	92	65	25
Term gestation	75	35-44	33
4-6 months	60	≈23	37
12 months		26-30	
Puberty	≈60	20	40
Adult	50-60	20	40

Alterations in body water compartment sizes will affect the volume of distribution of a drug. Total body water varies inversely with the amount of fat tissue in the body, due to the low water content of fat. Additionally, the extracellular fluid volume of newborn infants correlated more closely with bodyweight than with gestational age. In order to achieve comparable plasma and tissue concentrations of drugs distributing into the extracellular fluid, higher doses per kilogram bodyweight such as aminoglycosides must be given to infants and children than to adults.

Plasma protein binding. It is characterized by capacity and strength of binding and the rate of dissociation (K_a). Albumin, the major drug binding protein in plasma, binds primarily to acidic drugs but basic drugs bind more avidly to α_1-acid glycoprotein and lipoprotein than to albumin. Although serum albumin concentrations in neonate appear to be equivalent to adult values, α_1-acid glycoprotein concentrations are lower. The characteristics of protein binding during different stages of childhood are depicted in the table below(77).

Table 29 Physiological variables influencing plasma protein binding and drug distribution in infancy and childhood relative to adult values.

Parameter	Patient age group		
	Neonate	**Infant**	**Child**
Physiologic alteration			
Total protein	Decreased	Decreased	Equivalent
Plasma albumin	Decreased	Equivalent	Equivalent
Fetal albumin	Present	Absent	Absent
Plasma globulin	Decreased	Decreased	Equivalent
Free fatty acids	Increased	Equivalent	Equivalent
Unconjugated bilirubin	Increased	Equivalent	Equivalent
Blood pH	Low (7.1-7.3)	Equivalent (7.4)	Equivalent (7.4)
α_1-acid glycoprotein	Decreased	Data not available	Equivalent
Adipose tissue	Scarce (↑CNS)	Reduced	Generally reduced
Possible pharmacokinetic consequences			
Free fraction	Increased	Increased	Slightly increased
Apparent Vd			
Hydrophilic drugs	Increased	Increased	Slightly increased
Hydrophobic drugs	Reduced	Reduced	Slightly decreased
Tissue/plasma ratio	Increased	Increased	Slightly increased

Pathophysiological conditions which alter the extent to which a drug is bound to plasma or tissue proteins are thus capable of modifying both distribution and pharmacodynamic effects of the drug.

Table 30. Pathophysiologic factors that may affect drug-protein binding in children

Acidosis associated with hypoxemia
Malnutrition (hypoproteinemia)
Liver disease
Renal disease
Cystic fibrosis
Burns
Malignant neoplasms
Surgery
Trauma

Moreover, persistence of fetal serum proteins, hypoproteinemia (especially in premature neonates), the presence of competing ligands, and developmentally unique albumin-globulin interactions are age-related alterations in plasma protein (76). In summary, many of the age-related differences in drug distribution occur only during the first 10 to 12 months of life; thereafter, drug distribution in children is similar to that in adults (59-62, 76-77).

Blood pH. Neonates have a slightly lower pH (7.26-7.29) during the first few days of life compared to adults (7.35-7.45). Any decrease will render weak acids more dissociated from their binding sites, and thus the ratio of unbound to bound drug changes in favour of the unbound moiety. Thus, the frequently observed metabolic or respiratory acidosis of the neonate and, especially, of the premature infant may be associated with decreases in the binding of weak acid compounds to their plasma proteins.

Drug metabolism

Drug biotransformation occurs mostly in the liver and involves a series of metabolic reactions defined as phase I reactions (oxidation, reduction, and hydrolysis) that lead to either inactive or active compounds which may have pharmacologically activity equal, inferior or superior to parent compound and phase II reaction s

(conjugation with glucuronic acid and/or glycine and sulphate formation) that lead to inactive compounds.

Phase I reactions. The important factor that controls this biotransformation is the mixed function oxidase (MFO) system (cytochrome P-450, cytochrome b_5, NADPH cytochrome reductase. In both preterm and full term neonates, phase I reactions are significant decreased. Cytochrome P-450 activity in the fetus and in both premature and full-term neonate is approximately 50-70% of reported adult values (59-64, 76-77). The total P450 content was shown to remain stable from the first trimester of gestation to 1 year of age and account for about 30-50 % of the adult level (59-64, 76-77). Adult levels were 0.3±0.037 nmol/mg protein whereas levels in neonates and infants ranged from 0.08-0.13 nmol/mg/protein (64). NADPH cytochrome c reductase activity is lower in the premature than the full-term neonate, and both are much lower than adult values (63-64). With both enzymes, there seems to be a relationship between fetal and postnatal age and increasing enzyme activity.

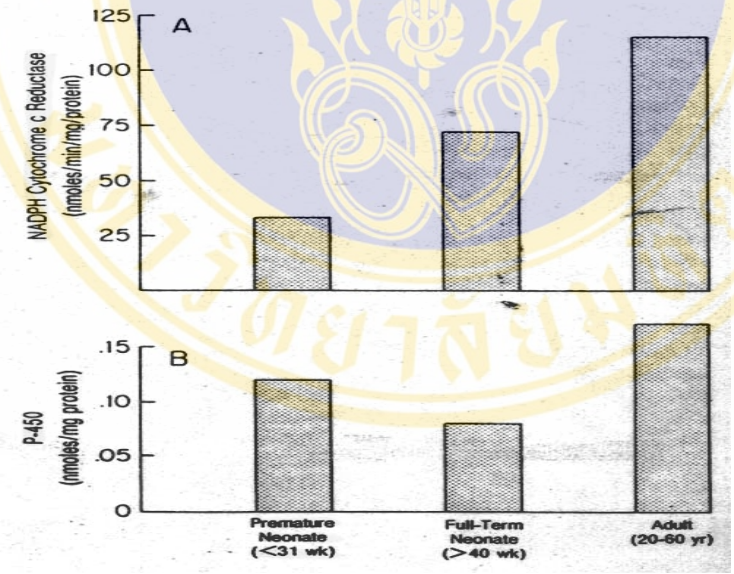

Figure 7. The extent of (A) NADPH cytochrome c reductase and (B) cytochrome P-450 from liver microsomal preparations of premature and full-term neonates and adult.

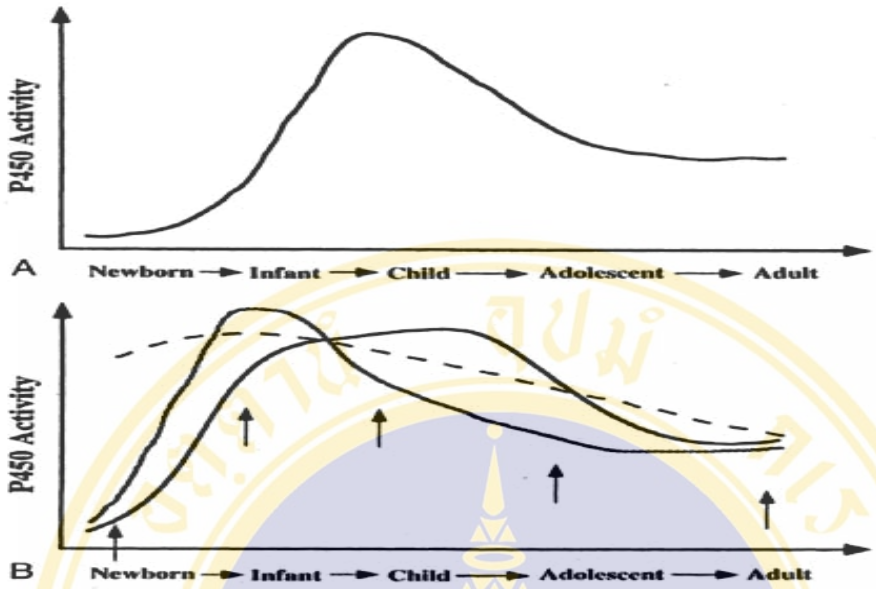

Figure 8. A. Traditional view of cytochrome P450 development. The development of functional P450 activity is traditionally viewed as being limited in newborn infants but increasing in the first year of life to levels in toddlers and older children that generally exceed adult capacity. Near the onset of puberty, oxidative drug biotransformation begins the decline to adult levels. B. Developmental profiles of hypothetical cytochromes P450. Not all P450s share the "traditional" developmental profile. Overall drug biotransformation capacity represents a composite of the individual drug metabolism pathways and is dependent on the isoforms and amounts of P450s (and other drug metabolizing enzymes) expressed. Therefore, for a given pediatric patient, the apparent drug metabolism "phenotype" is a function of that individual's unique complement of drug metabolizing enzymes and stage of development (vertical arrows).

The data from urinary excretion of metabolites of a variety of drugs showed that a low capacity for oxidative drug metabolism in full-term newborns and virtually none in premature infants (63-64). Similarly, the biological half-lives of drugs metabolized by the cytochrome P-450-dependent monooxygenase system are generally longer in infants than in adults (59-64, 76-77). N-demethylation is present in neonates, although in a reduced capacity compared with adults (63-64).

Microsomal epoxide hydrolase

Microsomal epoxide hydrolase is an enzyme that catalyses the hydrolysis of a large number of epoxide intermediates, which arise frequently from the oxidation of pharmaceutical and environmental compounds by the cytochrome P450 mixed function oxygenase system. The hepatic protein levels and enzyme activity were strongly conjugated with increasing gestational age. Fetal liver activities measured after approximately 4 months of gestation approached levels of approximately one-half of those in adult livers surveyed (63-64). No information about newborn children is available.

Alcohol dehydrogenase (ADH)

ADH is the rate-limiting enzyme responsible for the biotransformation of ethanol. The early pharmacokinetic data for ethanol in neonates support reduced clearance and enhanced accumulation in plasma; however, the plasma clearance was more rapid than adults and also to proceed by a first-order process (76). It might imply either a much lower Km or higher Vmax value value for ADH in young children or, alternatively, suggest a quantitative importance for other enzymes such as aldehyde dehydrogenase that contribute to ethanol metabolism (76). In addition, the elimination half-life for trichloroethanol in children between 1 and 13 years of age was significantly lower than preterm and term neonates (76-77). It would appear that by 12 to 30 months of postnatal age, ADH activity equal to or greater than that observed in adults is reached (76).

Phase II reactions. Conjugation reactions are the most important process. Glucuronidation is the most common conjugation pathway, due to the availability of glucuronic acid and the variety of functional groups with which it can combine. Glucuronide formation is catalyzed by a group of microsomal glucuronyl transferases, with uridine diphosphate-glucuronic acid as the donor of the glucuronide group. Some neonates are unable to conjugate bilirubin with glucuronic acid (64). Consequently, the increased unconjugated bilirubin can diffuse across the blood-ccumulate in brain tissue, potentially causing kernicterus. Glucuronide formation reaches adult values between the third and fourth year of life (59-64, 76-77).

Sulphation mainly involves phenolic hydroxyl group. It appears to be highly active in early gestation. Sulphation of acetaminophen is an important conjugation pathway instead of conjugation pathway in the full term neonate (76-77). Premature

neonates acetylate drugs such as sulfonamides, more slowly than do full-term neonates, and both are significantly lower than adult values (59-62, 76-77).

Conjugation of the aromatic carboxyl group with the α amino acids (mainly glycine) has been reported to be present in neonates at birth and reaches adult value around the sixth month of age (59-64, 76-77).

Table 31 Developmental pattern of phase I and phase II enzymes.

Enzyme(s)	Known Developmental Pattern
Phase I Enzymes	
CYP2D6	Low to absent in fetal liver but uniformly present at 1 week of postnatal age. Poor activity (approximately 20% of that in adults) at 1 month of postnatal age. Adult competence attained by approximately 3-5 years of age.
CYP2C19, CYP2C9	Not apparent in fetal liver. Inferential data using phenytoin disposition as a nonspecific pharmacologic probe suggest low activity in first week of life, with adult activity reached by 6 months of age. Peak activity (as reflected by average values for $V_{max,}$ which are 1.5-1.8 fold adult values) may be reached at 3-4 years of age which declines to adult values at the conclusion of puberty.
CYP1A2	Not present to an appreciable extent in human fetal liver. Adult levels reached by 4 months of age and may be exceeded in children 1-2 years of age. Activity slowly declines to adult levels, which are attained at the conclusion of puberty. Gender differences in activity are possible during puberty.
CYP3A7	Functional activity in fetus is approximately 30% to 75% of adult levels of CYP3A4.
CYP3A4	Low activity in the first month of life, with approach toward adult levels by 6-12 months of postnatal age. Pharmacokinetic data for CYP3A4 substrates suggest that adult activity may be exceeded between 1-4 years of age. Activity then progressively declines, reaching adult levels at the conclusion of puberty.
Phase II Enzymes	
NAT2	Some fetal activity present by 16 weeks. Virtually 100% of infants between birth and 2 months of age exhibit the slow metabolizer phenotype. Adult phenotype distribution reached by 4-6 months of postnatal age, with adult activity present by approximately 1-3 years of age.
TPMT	Levels in fetal liver are approximately 30% of those in adult liver. In newborn infants, activity is approximately 50% higher than in adults, with a phenotype distribution that parallels that in adults. In Korean children, adult activity appears at approximately 7-9 years of age.
UGT	Ontogeny is isoform specific as reflected by pharmacokinetic data for certain pharmacologic substrates (e.g. acetaminophen or chloramphenicol). In general, adult activity as reflected from pharmacokinetic data seems to be achieved by 6-18 months of age.
ST	Ontogeny (based on pharmacokinetic studies) seem to be more rapid than that for UGT; however, it is substrate specific. Activity for some isoforms (e.g. that responsible for acetaminophen metabolism) may exceed adult levels during infancy and early childhood.

We can summarize the activity and level of each enzyme during each stage of life until adult in the following table

Table 32 Summary of the ontogeny of phase I and enzymes (76-77).

Enzyme		Neonate	Infant	Child	Adult	Adult level
CYP1A1	activity	?	?	?	-	?
	mRNA	?	?	?	+	
	protein	+	?	?	?	
CYP1A2	activity	+/-	+	+++	+++	At 5-6 months
	mRNA	-	+	+++	+++	
	protein	-	+	+++	+++	
CYP1B1	activity	+	+	+	+	?
	mRNA	?	?	?	?	
	protein	?	?	?	?	
CYP2A6	activity	?	+	++	++	6-13 years
	mRNA	?	+	++	++	
	protein	?	+	++	++	
CYP2A13	activity	?	?	?	- (liver)	?
	mRNA	?	?	?	- (liver)	
	protein	?	?	?	-	
CYP2B6	activity	?	++	?	+	Higher levels found
CYP2B7	mRNA	?	++	?	+	in infant than adult
	protein	?	++	?	+	(n=1)
CYP2C	mRNA	++	++	+++	+++	30% of adult level
	protein	+	++	+++	+++	from 1st wks until 1 yr
CYP2C8	activity	?	?	?	?	
CYP2C9	activity	+	+	++	++	Increasing in 1st week to 50% and adult levels not before 1 year
CYP2C18	activity	?	?	?	?	
CYP2C19	activity	+	+	++	++	9 months
CYP2D6	activity	+/-	+	+	+	
	mRNA	++	+	+	+	
	protein	+/-	+	+	+	
CYP2E1	activity	+	+	++	++	Protein levels after 9
	mRNA	+/-	+	++	++	months
	protein	+	+	++	++	
CYP3A4	activity	+/-	++	+++	+++	In the first
	mRNA	?	++	+++	+++	year/replacing 3A7
	protein	?	++	+++	+++	
CYP3A5	activity	?	?	?	?	?
	mRNA	+	+	++	++	
	protein	+	+	++	++	
CYP3A7	activity	++	+	-	-	Activity is high in
	mRNA	?	?	?	?	fetus, with a peak in
	protein	?	?	?	?	first week after birth and decrease in first year

Table 33 Summary of the ontogeny of Phase II enzymes (76,93).

Enzyme		Neonate	Infant	Child	Adult	Adult level	Effect on clearance
UGT1A1	activity mRNA protein	+/- ? +/-	+ ? +	+ ? +	+ ? +	3-6 months	
UGT1A3	activity mRNA protein	? ? +	? ? ?	? ? ?	- + ?	?	
NAT2	activity mRNA protein	+/- ? ?	+/- ? ?	+ ? ?	+ ? ?	10-12 months	
Sulfotransferase	activity mRNA protein	+ ? ?	+ ? ?	+ ? ?	+ ? ?	From birth	
TMPT	activity mRNA protein	++ ? ?	? ? ?	? ? ?	+ ? ?	50% higher in neonates than adults	

Gene	Prenatal Trimester			Neonate	1 Month to 1 Year	1 to 10 Years	Adult
	1	2	3				
GSTA1/A2	+	+	+	+	+	+	+
GSTM	+	+	+	+	+	+	+
GSTP1	+	+	+	+	-	-	-
NAT2	+	+	+	+	+	+	+
UGT1A1	-	-	-	+	+	+	+
UGT1A3	?	+	+	+	+	+	+
UGT1A6	-	-	-	+	+	+	+
UGT2B7	?	+	+	+	+	+	+
UGT2B17	?	+	+	+	+	+	+
EPHX1	+	+	+	+	+	+	+
EPHX2	?	+	+	+	+	+	+
SULT1A1	?	+	+	+	+	+	+
SULT1A3	?	+	+	+	+	+	+
SULT2A1	-	-	+	+	+	+	+

+, activity or protein detectable; -, activity or protein not detectable; ?, not determined.

Table 34 Postulated ontogeny of human uridine 5′ - diphosphate glucuronosyltransferase (UGT) (93)

UGT isoform	Activity	
	In vitro[a]	In vivo[b]
1A1	Nearly undetectable during fetal life, increasing directly after birth, reaching adult levels 3 to 6 months after birth	NA
1A3	During fetal and neonatal period, around 30% of adult activity	NA
1A6	Glucuronidation during fetal life at 1 to 10% of adult levels, increasing slowly after birth, at 6 months 50% of adult activity. Independent of gestational age.	Not reaching adult levels before age 10 years
1A9	NA	NA
1A10	NA	NA
2B4	NA	NA
2B7	15 to 27 weeks fetal life: 10 to 20% of adult levels. Change in activity regulated by birth-related events?	First 2 weeks of life 20% of levels of older (1 to 16 years) children. No apparent change during adolescence
2B15	NA	NA
2B17	Reduced activity during fetal (<10%) and neonatal (around 10%) period when compared with adult	NA

a Data from immunoreactivity or catalytic studies with fetal, neonatal or adult human liver microsomes using UGT substrates that are mainly metabolized by 1 specific UGT isoform.
b Data derived from pharmacokinetic studies using UGT substrates that are mainly metabolized by 1 specific UGT isoform.
NA = no information available.

Table 35 Metabolic pathway differences between children and adults.

Reaction	Pediatrics	Conditions
Reduction	Equivalent	
Hydroxylation	~20-70% adult level	
Dealkylation	Decreased	
Sulfation	Equivalent	>in 7-10 year olds than adults
Glycine conjugations	Equivalent	
Glucuronidation	Decreased	Compensated for by sulfation
Esterification	Decreased	~10-12 months adult rate reached
N-demethylation	Decreased	
Methylation	Increased	
UDPG-glucuronyltransferase	Decreased	Adult rate reached by 3 years old
Alcohol dehydrogenase	Decreased	Adult rate reached by 5 years old

In conclusion, the capacity for drug biotransformation in both premature and full-term neonate during the first two weeks of life is reduced (59-62, 76-77, 93). However, for most drugs this is not a serious problem since a reduction in the primary metabolic pathway is often compensated for by increased activity of other pathways (93).

The reduced metabolic activity in the neonate is probably the result of the following factors: decreased cytochrome P_{450}, decreased NADPH cytochrome-C-reductase, the presence of endogenous inhibitors of maternal origin, reduced liver blood flow and relative hypoxaemia. Nevertheless, both premature and full-term neonates metabolise drugs at a rate which is several times lower than in adult. This is generally present during the first 2 to 3 weeks of extrauterine life and is then followed by a dramatic increase in the rate of phase I reactions, which in 5 to 6 days may change so that clearances increase from about 20% of those in adults to 2 to 4 times greater (63-64, 76-77, 93).

Drug metabolism in the lumen of the gastrointestinal tract

At birth, the enterobacteria and enterococci are the predominant microflora of the human gastrointestinal tract, whereas bifidobacteria are absent but appear after 7 days and become the predominant flora after 13 days (76). In some instances, the lack

of xenobiotic metabolizing ability observed is not due to absence of certain microflora but rather to immaturity of the bacterial enzyme systems in the gut lumen (76-77).

Table 36. Ontogeny of faecal bacterial enzymes in healthy infants aged 0-6 months

Nmoles/mg protein × min	0 months (n=29)		1-2 months (n=27)		6 months (n=26)	
	Mean	Range	Mean	Range	Mean	Range
Urease	0.9	0-12.4	0.4	0-4.6	10.7	0-48.1
β-glucosidase	1.1	0-4.1	2.1	0-9.9	7.9	0-82.8
β-glucuronidase	0.7	0-5.4	0.3	0-1.5	0.9	0.04-8.0

The appearance of bacterial enzymes in faeces after birth was slow for urease but more rapid for β-glucosidase and β-glucuronidase. At 6 months of age, nearly all infants harboured β-glucosidase and β-glucuronidase, while urease was detected in only 67% of the infants (76). Urease and β-glucosidase activities increased significantly during the 6-months follow-up period, while the β-glucuronidase activity did not change during the study period (76).

Excretion

The immaturity and postnatal development of renal elimination processes have important implications for drug disposition . First, the neonatal kidney is inefficient at drug elimination, which leads to prolonged elimination half-lives. Second, the maturation of renal drug-elimination processes occurs at variable rates and may be influenced by a number of factors such as maternal and postnatal exposure to drugs. Finally, the mechanisms of uptake and storage in renal tubular cells are subject to maturational changes that may lead to age-related differences in intrarenal accumulation of drugs.

Compared with the full-term neonate, the premature neonate is born with a deficient number of glomeruli (59-62, 65, 67, 76-77). Additionally, mean renal clearance in neonates less than one week of age was 32.9±7.4 ml/min/1.73m^2, increasing to 88.9±23.9 and 144.4±38.4 ml/min/1.73m^2 at three months and 1.5 years, respectively 965, 67). Premature neonates are reported to demonstrate longer half-lives (56-88 hours) than their full-term counterparts (59-62, 65, 67, 76-77).

Renal blood flow

Renal blood flow and renal plasma flow increase with age as a result of an increase in cardiac output and a reduction in peripheral vascular resistance. The kidneys of a neonate only receive 5 to 6% of total cardiac output compared with 15 to 25% for adults. Renal plasma flow averages 12 ml/min (0.72 l/hr) at birth and increase to 140 ml/min (8.4 l/hr) by 1 year of age (65, 67). If renal plasma flow is corrected for body surface area, adult values are reached before 30 weeks of extrauterine life (65, 67). Moreover, renal blood flow appears to increase in proportion to the development of the renal tubules (59-62, 65, 67, 76-77).

Glomerular filtration (GFR)

GFR is correlated with gestational age but this correlation is not linear in neonate prior to 34 weeks gestation (59-62, 65, 67, 76-77). GFR is lower in neonates born before 34 weeks' gestation compared with full-term neonates (0.6-0.8 ml/min vs 2-4 ml/min respectively) (65, 67). GFR, as measured by creatinine clearance or clearance of inulin remains relatively constant at low rates of 1 ml/min prior to 34 weeks of gestation (65, 67). The reason for this appears to correlate with the ontogeny and functional organization of the glomerulus. Premature neonates develop their GFR more slowly than that of full-term neonates during the first week of life (59-62, 65, 67, 76-77). In the first 2 to 3 days of postnatal life, there is a marked increase in GFR of full term babies to rates between 8 and 20 ml/min, compared with increases in neonates less than 34 weeks gestation to 2 to 3 ml/min (59-62, 65, 67, 76-77). The increase in GFR after birth has been shown to be dependent on postconceptual age, and not postnatal age. Multivariate analysis indicated that gestational age, but not body weight, was the major determinant for the increase in mean GFR (67). The postnatal increase in GFR is due to the increase in cardiac output which is associated with specific changes in renal vascular resistances, changes in renal blood flow distribution, and a higher permeability of the glomerular membrane. Moreover, there is a glomerular/tubular imbalance ; the more advanced maturation of glomerular function as compared with proximal tubular secretion (67). This imbalance may persist up to 6 to 10 months of age, at which time both tubular and glomerular function approach values approximately equal to those observed in healthy young adults (67).

Tubular function

This process is an active process that requires both energy and a protein carrier, consists of separate system for organic acids and organic bases. Tubular secretory function of bicarbonates, phosphates, glucose and para-amino hippurate are reduced in neonates compared with adult (65, 67, 76-77). This reduction may be due to decreased blood flow to peritubular regions, short tubules with thickened epithelium, immature energy-supplying processes, and insufficient carrier mechanisms.

Proximal convoluted tubules in the normal kidney of a full term infant are small in relation to their corresponding glomeruli (65, 67). This imbalance in size is reflected by functional differences in the transport capacity (secretion) of the proximal tubular cells. Using the tubular transport maximum for para-amino hippurate (TmPAH), a compound secreted by the proximal tubules, as an indicator of tubular function, there was a 10-fold increase in TmPAH in the first year of life, with adult values being reached by 30 weeks of life (65, 67). Consequently, tubular function matures at a slower rate than glomerular function (59-62, 65, 67, 76-77). Reasons for this reduction include not only the small size of the tubules, but also a smaller mass of functioning tubular cells, reduced blood flow to the outer cortex, and immaturity of energy-supplying processes (59-62, 65, 67, 76-77).

Tubular reabsorption of drugs consists of either passive diffusion or active transport. Passive diffusion is the more common process and it is decreased in neonates. The reduction in transport capacity dues to the low blood flow in peritubular regions, the immaturity of energy-supplying processes, the small mass of working tubular cells, and the small size of undeveloped tubuli.

Clearance

Intrinsic clearance can be defined as the maximal ability of the organ to remove the unbound drug or it shows the intrinsic capacity of hepatic enzymes (59-62, 65, 67, 76-77). A reduction of intrinsic clearance may have a dramatic effect for drug with low extraction, such as diazepam, theophylline, salicylate and phenobarbitone. A lower intrinsic clearance is synonymous with a slower production of metabolites so a greater probability of accumulation of the parent drug in the body.

In the human neonate, either premature or full term, all drug elimination processes are significantly reduced (59-62, 65, 67, 76-77). The metabolism and renal

excretion of many drugs are initially severely impaired, but show a rapid improvement in the first month of life. However, this rapid acquisition of developmental competence in drug clearance in the first month of life is strongly dependent on maturational and drug-induced changes in renal function of the preterm infant (59-62, 65, 67, 76-77). The following table reviews the differences in drug disposition between newborns and adults.

Table 37 Drug disposition in infants compared with that in adults: potential influence of pharmacokinetics

Disposition parameter	**Newborn vs adult**	**Possible pharmacokinetic result**	**Example drug**
Absorption	↓	↓ AUC	Penicillins, sulphonamides
Volume of distribution	↑	↓ Peak	Gentamicin, digoxin
% Protein binding	↓	↑ Free fraction	Clindamycin, theophylline
Metabolism	↓	↓ Clearance	Chloramphenicol, theophylline
Excretion	↓	↑ AUC ↑ $t_{1/2\,\beta}$	Gentamicin, furosemide
Abbreviation: ↓ = less than in newborns than in adults; ↑ = greater in newborns than in adults; AUC = area under the concentration-time curve; $t_{1/2\,\beta}$ = elimination half-life.			

From the report by Bueva et al (67) about renal function in 66 preterm and term neonates, the result could be summarized as following table.

Table 38 Plasma creatinine, sodium concentration, and creatinine clearance during the first week of life (mean values±SEM)

	Postnatal days			
Group	**1-2**	**8-9**	**15-16**	**22-23**
I (1001-1500 g)				
Cr (μmol/l)	95±5	64±5	49±4	35±3
Na (mmol/l)	137±1	135±1	132±2	136±2
CrCL (ml/min)	0.65±0.14	1.31±0.24	1.73±0.29	
n	11	10	8	8
II (1501-2000 g)				
Cr (μmol/l)	90±5	58±7	50±8	30±2
Na (mmol/l)	136±2	139±1	131±2	139±1
CrCL (ml/min)	0.92±0.19	1.91±0.24	2.86±0.56	
N	15	11	11	9
III (2001-2500 g)				
Cr (μmol/l)	83±5	47±8	38±8	30±10
Na (mmol/l)	134±2	138±1	135±2	136±3
CrCL (ml/min)	1.42±0.31	3.1±0.6	3.84±1.3	
N	12	7	4	2
IV (full-term)				
Cr (μmol/l)	66±3	40±4	30±8	27±7
Na (mmol/l)	136±1	139±1	137±1	135±3
CrCL (ml/min)	3.36±0.32	5.17±0.93	7.52±1.9	
n	28	14	4	3

Plasma creatinine concentration. These data indicated that the plasma creatinine of newborn infants are elevated at birth and the concentration is significantly more elevated in the low birth weight infant, with values of 66±3 μmol/l in the term infants (group 4) compared with95±5 , 90±5, and 83±5 μmol/l in group 1, 2,and 3, respectively (67). Moreover, it decreases rapidly to reach stable levels within 3 weeks (67). Interestingly enough, the plasma creatinine is significantly higher in very low birthweight infants and appears to be inversely related to gestational age ($r = -0.738$ to -0.795; $p<0.001$). The elevation probably reflect the difficulty these very low birthweight infants have eliminating the excess creatinine transferred from the mother: their GFR is still too low during the first postnatal days to effectively eliminate this excess.

Plasma sodium concentration. The result showed that both term and preterm neonates are able to maintain sodium balance on a standard sodium intake of 1-2 mmol/kg/day during the first weeks of life although it has been claimed that premature

neonates have difficulties in maintaining sodium balance when administered the same amount of sodium intake because of the deficient proximal reabsorption and incapacity of the distal tubule to deliver an increased fractional sodium load despite high serum aldosterone levels.

Creatinine clearance. Creatinine clearance was low at birth; the lowest values were observed in infants with the lowest birthweight. It increased in the following three weeks in all infants and it correlated significantly with postnatal age. Additionally, the velocity of the postnatal maturation of creatinine clearance was higher in term neonates ($p<0.01$) both when it was expressed in absolute terms and when it was factored by the body surface area.

Cockcroft-Gault equation, widely used to calculate creatinine clearance, is not applicable and not accurate in neonates because creatinine clearance may overestimate GFR at low GFR, neonate's creatinine is not accurate at birth (mother's creatinine), and little muscle mass levels to low creatinine (lab inaccuracy at low concentration). However, in children, creatinine clearance may be used but creatinine value must be stable and patient must not be emaciated.

There are many factors that may have a considerable impact on drug kinetics and effects in the neonate and the young infant. For example, hypoxaemia, renal and gastrointestinal pathology, cardiac insufficiency, respiratory distress syndrome, and modification of the regular haemodynamic development.

Patent ductus arteriosus (PDA) infants is associated with a variety of physiologic changes that could alter drug disposition (66). Perfusion of drug-elimination organs may be diminished, resulting in decreased drug elimination (66). The general fluid overload state associated with PDA may result in larger volumes of distribution (V_d), and dilutional effects for many drugs (66). The presence of acidosis may also increased protein binding of some drugs (e.g. theophylline) and consequently increase V_d (66). Concurrent acidosis may alter the ionization of agents with a pK_a close to 7.4 such as Phenobarbital, permitting increased concentrations of unionized molecules that are available to cross biologic membrane more freely and potentially distribute more extensively into tissue (66). These changes in condition can be summarized in the table 39.

Table 39 Physiologic factors associated with patent ductus arteriosus that may alter drug disposition.

Clinical status	Physiologic change	Pharmacokinetics impact
Hypoxia	Gastrointestinal hypoxia or hypoperfusion	Absorption ↓ bioavailability
Poor nutrition	↓ protein / albumin	Protein binding ↑ unbound drug
Acidosis (↓pH)	↓ protein binding ↑ unionized drug (for weak acids) →↑ tissue distribution	Distribution ↑ volume of distribution
Fluid overload /edema	↑ intracellular and extracellular water	
Left to right blood shunt through PDA (to lungs)	Creation of a third compartment in pulmonary vasculature	
Blood shunt through PDA	↓ cardiac output →	Elimination
Hypoxia	↓ liver blood flow	↑half-life
Congestive heart failure	↓ renal blood flow	↓clearance
Indomethacin therapy	↓ renal blood flow	
PDA = Patent Ductus Arteriosus; ↓ = decreased; ↑ = increased.		

In summary, premature infants have many differences in physiologic factors such as drug metabolism organs and function from younger children and adult. Additionally, they are prone to many disease or disorder that may affect the pharmacokinetics of many drug. Drug utilization in these groups should be very aware of overdosing that may be more harmful than the older children and adult. Therapeutic drug monitoring in high alert drugs are necessary.

CHAPTER 3

MATERIALS AND METHODS

Study Design

The study was designed as prospective randomized controlled trial study for evaluating the efficacy of ibuprofen suspension (Junifen®) in prevention of PDA compared with placebo. Additionally, the following terms were used throughout the study period:

1. PDA (Patent Ductus Arteriosus) is the condition that patient's ductus arteriosus is still opening by echocardiogram.

2. **Silent PDA** is refer to the patient with echocardiographic evidence of left-to-right ductal shunting but without any signs of patent ductus arteriosus.

3. **IVH (Intraventricular Hemorrhage)** is an intracranial hemorrhage usually arising in the germinal matrix and periventricular regions of the brain determined by cranial ultrasound. It is grouped into four separate grades (17).

Grade I – subependymal hemorrhage : There are bilateral germinal matrix (subependymal) hemorrhages that not ruptured into the ventricles.

Grade II – intraventricular hemorrhage without ventricular dilatation : There are intraventricular extravasation in addition to bilateral germinal matrix hemorrhage.

Grade III - intraventricular hemorrhage with ventricular dilatation : There are progressive dilatation of the ventricles such as the body and the occipital horn of the lateral ventricles are filled with blood and are dilated.

Grade IV – intraventricular hemorrhage with parenchymal hemorrhage : There is an extension of hemorrhage into the parenchymal of cerebral hemisphere. It may have

progressive ventricular enlargement and a periventricular cyst at the site of the parenchymal hemorrhage.

4. **ROP (Retinopathy of Prematurity)** is the ocular underdevelopment due to interuption of normal progression of newly forming retinal vessels. Vasoconstriction and obliteration of the advancing capillary bed is followed in succession by neovascularization extending into the vitreous, retinal edema, retinal hemorrhages, fibrous and traction on, and eventual detachment of, the retina. It is staged according to the international classification (18).

Stage I – Demarcation line : There are recognizable abnormal branching or arcading of vessels leading up to the thin line that separates the avascular retina anteriorly from the vascularized retina posteriorly. The line is relatively flat and white and lies within the plane of the retinas.

Stage II – Ridge : It is the growing of line of stage I that occupies a volume, and extends up out of the plane of the retina to enter it.

Stage III – Ridge with extraretinal fibrovascular proliferation : The ridge of stage II is added the pressence of extraretinal fibrovascular , and proliferative tissue.

Stage IV – Retinal detachment: There are unequivocal detachment of the retina, It may be caused by an exudative effusion of fluid, traction , or both.

5. **BPD (Bronchopulmonary Dysplasia)** is the chronic pulmonary condition of infants who have experienced respiratory failure, remain oxygen dependent for at least 28 days postnatal age and have chronic pulmonary pathologic disease manifested as areas of hyperinflation and atelectasis on chest radiograhs. It can be described by four radiographic stages.

Stage I - The radiographic features of hyaline memberane disease

Stage II - The findings of congestive heart failures and pulmonary edema from a PDA with a whiteout on the chest radiograph.

Stage III - Pulmonary hyperinflation and areas of emphysema and atelectasis

Stage IV - Large areas of emphysema and atelectasis with overall pulmonary hyperinflation. Morever, there is a gradual decrease in the degree of hyperinflation over a period of many years.

6. **NEC (Necrotizing Enterocolitis)** is an acquired neonatal disorder representing and end-expression of serious intestinal injury following a combination of vascular, mucosal, and toxic insults to relatively immature gut. It is classified into stages according to Bell et al (1978) as followings.

Stage I – Suspect

a. Any one or more historical factors producing perinatal stress.
b. Systemic manifestations include temperature instability, lethargy, apnea, bradycardia.
c. GI manifestations include poor feeding, increasing pregavage residuals, emesis (may be bilious or test positive for occult blood), mild abdominal distension; occult blood may be present in stool (no fissure).
d. Abdominal radiographs show distension with mild ileus.

Stage II – Definite

a. Any one or more historical factors.
b. Above signs and symptoms plus persistent occult or gross GI bleeding ; marked abdominal distension.
c. Abominal radiographs show significant intestinal distension with ileus; small bowel separation (edema in bowel wall or peritoneal fluid), unchanging or persistent “rigid” bowel loops, pneumatosis intestinalis, portal vein gas.

Stage III – Advanced

a. Any one or more historical factors.
b. Above signs and symptoms plus deterioration of vital signs, evidence of septic shock, or marked GI hemorrhage.
c. Abodominal radiographs may show pneumoperitoneum in additon to others listed in IIc.

7. **Pharmacokinetic** is the quantitative relationship between administered doses and dosing regimens and (observed) plasma and/or tissue concentrations of the drug.

8. **Pharmacodynamic** is defined as the quantitative relationship between (observed) plasma and/or tissue concentration(s) of the active moiety and the magnitude of the (observed) pharmacological effect(s).

9. **Pharmacokinetic / Pharmacodynamic model** is termed as a mathematical description of these relationships; the model parameters provide information about intrinsic drug properties. The knowledge of the combined Pharmacokinetic / Pharmacodynamic model and the parameter estimates allows prediction of concentration vs time (C-t) and effect vs time (E-t) profiles for different dosing regimens.

10. **Drug Absorption** is defined as the process by which the unchanged drug proceeds from the site of administration to the site of measurement within the body.

11. **Drug Distribution** is the process of reversible transfer of a drug to and from the site of measurement, usually the blood or plasma.

12. **Drug Metabolism** refers to the process by which drugs are biochemically modified to facilitate their degradation and subsequent removal from the body.

13. **Drug Elimination** is the irreversible loss of drug from the site of measurement. It occurs by two processes, excretion and metabolism.Firstly, excretion is the irreversible loss of the chemically unchanged drug. Lastly, metabolism is the conversion of one species to another.

14. **Cmax (Maximun concentration)** is defined as the height of the peak of the concentration-time curve of a drug in blood/plasma/serum represents the maximum concentration of the drug achieved after its administration. It determines the therapeutic efficacy and toxicity of the drug.

15 **Tmax (Time to maximum concentration)** is the time elapsed between drug administration and achieving Cmax or it is the time required to achieve Cmax.

16. **AUC (Area under the curve)** represents the area under the blood, plasma or serum concentration-time curve. It reflects the total amount of drug in the body following the administration of a drug after a specific dose. It is measured mathematically, usually employing a technique known as “trapezoidal rule”.

17. **F (Bioavailability)** is defined as the fraction of unchanged drug that reaches the systemic circulation after the administration of a drug dose.

18. **V_d (Volume of distribution)** is a proportional constant that relates the amount of drug in the body to the concentration of drug in blood/plasma/serum. It provides an estimation of how widely a drug is distributed in the body/tissues.

19. **V_d/F (Apparent volume of distribution)** is the available volume in both the general circulation and the tissues of distribution of the body which would be required to contain the administered dose if that dose is evenly distributed at the concentration measured in plasma or blood. It gives a mathematical measure of the extent of tissue distribution, but does not give any anatomical or physiological information about that distribution. It may be estimated if the total dose given and the plasma drug concentration after equilibrium are known:

$$V_d/F = \frac{\text{Amount (mg) of drug in body}}{\text{Plasma drug concentration (mg/ml) after equilibration}}$$

20. **CL (Clearance)** refer to the volume of blood/plasma/serum that would be completely cleared of drug per unit time. It is the best parameter that describes the elimination capacity of the body.

21. **$t_{1/2}$ (Elimination half-life)** is the time taken for the concentration profile of a drug in blood/plasma/serum to fall by half. It is a secondary variable determined by volume of distribution and clearance.

Subject

1. Number of subjects recruitment

Sample size was calculated based on PDA closure that was closed and not closed (dichotomous response variables) after ibuprofen administration. These data are ordinal data with two groups (two independent samples) so the statistical formula (20) used was:

$$2N = \frac{4(Z_\alpha + Z_\beta)^2 \, \bar{p}\,(1 - \bar{p})}{D^2} \qquad \text{when } \bar{p} = \frac{P_1 + P_2}{2}$$

The following terms are defined as

$2N$ = total sample size (N participants / group)

Z_α = 1.96 for two-tailed $\alpha = 0.05$

Z_β = 0.84 for $\beta = 0.20$ (power 80%)

P_1 = the probability of PDA closure in intervention group

P_2 = the probability of PDA closure in control group

D = the difference of PDA closure rate between intervention group vs control group, $P_1 - P_2$

The previous study by Phatcharaphun et al (14) compared the efficacy of ibuprofen suspension in prophylaxis group with placebo group. The result showed that the prophylaxis group had higher PDA closure rate than control group (80.0% vs 34.0% ; P = 0.016). By using above statistical formula, the figure can be substituted.

$Z_\alpha = 1.96$ $Z_\beta = 0.84$

$P_1 = 0.80$ $P_2 = 0.34$

$D = P_1 - P_2 = 0.80 - 0.34 = 0.46$

$$\bar{p} = \frac{P_1 + P_2}{2} = \frac{0.80 + 0.34}{2} = 0.57$$

$$2N = \frac{4\,(1.96 + 0.84)^2\,(0.57)\,(1 - 0.57)}{(0.80 - 0.34)^2}$$

$2N = 36$; $N = 18$

By calculation we obtained 18 recruited patients in each group but we approximated 20% drop out rate so the actual number of recruited patients were 21 in each group. In conclusion, the total subjects recruited were 42 persons.

2. Randomization

We used blocked randomization method in order to balance the number of participant in each group during the course of randomization. There are two important advantages of this method. Firstly, if the type of participant recruited for the study changes during the study period, blocking will produce more comparable group. Lastly, if the trial should be terminated before enrollment is completed, balance will exist in terms of number of participants randomized to each group.

3. Population characteristics

Population that involved in this study are premature infants in neonatal unit and ICU ward at Queen Sirikit National Institute of Child Health (QSNICH) with the following characterisitics.

Inclusion criteria

1 Patients with gestational age ranged from 28-32 weeks and birth weight less than or equal to 1,500 grams.

Exclusion criteria

1 Patients with congenital heart defect (other than PDA).
2 Patients whose their mothers had evidence of overt prenatal infection, history of drug abuse.
3 Patients who have evidence of intrauterine infection, including toxoplasma, rubella, cytomegalovirus, herpes simplex (TORCH).
4 Patients who have a history of antenatal nonsteroidal anti-inflammatory drugs administration.
5 Patients who have persistent pulmonary hypertension.
6 “Nonviability” of infant including unstable clinical condition or moribund infant (dying in first 24 hours) or infant with contraindication to oral feeding.
7 Other major congenital anomaly, including chromosomal abnormalities.
8 Hydrops fetalis.
9 Patient with serum creatinine concentration is greater than or equal to 1.5 mg/dl, platelet count is less than or equal to 75,000 /μl, and presence of clinical manifestation of abnormal clotting function.
10 Inability to obtain informed consent.

Methods

1. Patient recruitment : We recruited patients who met the above inclusion criteria. Then the physician explained the details of this study to their parent. These included background, rational, objective, expected outcome, advantage, disadvantage,and method of the study. After their parent had understood and pleased to have their infants to enter the study, the informed consent were signed. Moreover, the pediatric cardiologist examined all recruited patients by echocardiogram within 24 hours to exclude other congenital heart defects.

2. Randomization : We assigned each recruited patient into control group or study group as order calculated previously by the blocked randomization method.

3. Laboratory assessment : We could summarize the laboratory examination schedule as the following table.

Table 40 Time schedule for laboratory examination in both study groups

Variables	Day0	Day1	Day2	Day3	Day4	Day5	Day6	Day7
Urine output (ml)	√	√	√	√	√	√	√	√
Fluid intake (ml)	√	√	√	√	√	√	√	√
Serum BUN (mg/dl)	√			√				
Serum creatinine (mg/dl)	√			√				
Electrolyte	√			√				
CBC, Coagulogram	√			√				

4. Drug administration and blood sampling : Nothing other than the medication was given per oral during the first three days.

Patients in study group were obtained umbilical catheter first. Then they received three doses of ibuprofen suspension (Junifen) 10 mg/kg via orogastric tube and followed by 0.5 ml water. The first dose was given within 24 hours after birth. The second and the third dose were given apart from the first dose 24 and 48 hours respectively. During each dose of drug administration, blood samples were drawn from umbilical catheter by two syringe method as the following time table

Table 41 Time schedule for blood sampling in study group.

Time	30 min before	before	2 hr after	4 hr after	8 hr after	10 hr after	12 hr after	14 hr after	18 hr after
First dose		0.2ml	0.2ml	0.2ml	0.2ml	0.2ml	0.2ml	0.2ml	0.2ml
Second dose	0.2ml				0.2ml				
Third dose	0.2ml				0.2ml				

The blood samples were used to analyze plasma drug concentration by High Performance Liquid Chromatography (HPLC) method. Drug level from HPLC analysis also used to calculate basic pharmacokinetic parameters in each patient.

The amount of drug given to preterm infants was calculated based on patient's birthweight. It could be summarized as following table.

Table 42 Amount of ibuprofen suspension administration measured by insulin syringe.

Body weight (gm)	Received Dose (mg)	Amount of drug label on insulin syringe#
800	8	40
850	8.5	43
900	9	45
950	9.5	48
1000	10	50
1050	10.5	53
1100	11	55
1150	11.5	58
1200	12	60
1250	12.5	63
1300	13	65
1350	13.5	68
1400	14	70
1450	14.5	73
1500	15	75

Ibuprofen suspension 1 teaspoonful (5ml) = 100 mg [1 ml = 20 mg]

Insulin syringe 1 ml has 100 points so 1 mg ibuprofen = 5 points insulin syringe

The control group received an orange starch suspension that looked like ibuprofen suspension (Junifen) with the same method and time schedule as the study group. In contrast, blood samplings were not performed after each given dose.Both ibuprofen suspension (Junifen) and orange starch suspension were filled in the same container, amber glass that didn't react with the drug or other ingredients.

5. PDA diagnosis : Evaluation for PDA was done by both clinical and echocardiographic examination as the following protocol

Clinical diagnosis of PDA include bounding pulse (pulse pressure > 35 mmHg), hyperactive precordium, tachycardia (heart rate > 170 beats/min), continuous murmur, hepatomegaly, cardiomegaly (cardiothoracic ratio > 0.6) and increased pulmonary vasculature on chest X-ray.

Echocardiographic examination includes the size of PDA, the existence of left atrial enlargement (LAE), left ventricular enlargement (LVE) and the evaluation of pulmonary pressure (PAP) if possible.

Table 43 Schedule for PDA evaluation by echocardiogram and clinical examination.

PDA evaluation by	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 14	Day 21	Day 28
Clinical presentation	√	√	√	√	√	√	√	√	√		√
Echocardiogram	√			√				√	√	√	√

6. Secondary outcome : These included presence of intraventricular hemorrhage, gastrointestinal function (feeding tolerance and necrotizing enterocolitis), respiratory status (PPHN and broncopulmonary dysplasia), duration of hospitalization, discharge age, and discharge status.

7. Rescue treatment : If the patient shows sign of PDA evaluated by echocardiogram or clinical finding after completing drug administration course, the standard indomethacin treatment will be performed as the following regimen.

Table 44 Dose of indomethacin for back up treatment.

Age at first dose	Dose (mg/kg)		
	First dose	Second dose	Third dose
< 48 hours	0.2	0.1	0.1
2 – 7 days	0.2	0.2	0.2
> 7 days	0.2	0.25	0.25

If the ductus arteriosus stills opening, the surgical ligation will be performed .

Problems that may be occurred during study and resolutions.

* If the patient vomits before 45 minutes after the first dose is given, we will delay time of drug administration until the patient can be fully feeded. In case of the patient vomits after 45 minutes after the first dose is given, the next drug administration time is not changed if the patient can be fully feeded.

* If the patients show any signs of serious side effect or adverse drug reaction that has not been reported from previous studies after or during the given drug, they will be excluded from the study. Then appropriate treatment will be performed immediately.

* Although there are less renal side effect of this drug, transient oliguria (urine output < 1 ml, BUN ≥ 30 and S_{cr} ≥ 1.5) after first dose or second dose may occur. If it does, the patient will be excluded from the study and they will be received appropriate treatment.

* If the patients show sign of NEC (poor feeding, increasing pregavage residual, emesis, abdominal distension,and stool occult blood), they will be excluded from the study and appropriate treatment will be performed immediately.

Analysis of drug by HPLC (High Performance Liquid Chromatography).

Material

Drugs, reagents, and chemical substances	Supplier
Ibuprofen suspension (Junifen®) Lot No. 306RR, Mfg Date : Oct 2002, Exp date : Oct 2005	The Boots Manufacturing Co.,Ltd. Thailand.
Meglumine Indomethacin (Liometacen®) (Meglumine indomethacin 77.2 mg equivalent to indomethacin 50 mg and excipients to 195.738 mg) Batch no 030065, Mfg date : Apr 2003, Exp date : Apr 2006	Chiesi Farmaceutici S.p.A. Parma, Italy. Distributor in Thailand: Pacific Healthcare (Thailand) Co; Ltd.
Water for Injection 2 ml (Solvent for Liometacen®) Batch no 030087, Mfg date : Apr 2003, Exp date : Apr 2006	
Standard ibuprofen powder Batch no IBU0211845, Mfg date : Nov 2002, Exp date : Oct 2007, Assay 99.6%	Siam Pharmaceutical Co.,Ltd. Thailand
Standard mefenamic acid powder Lot no 020905, Mfg date : Sep 2002, Exp date sep 2006, Assay 99.8%	Siam Pharmaceutical Co.,Ltd. Thailand
Methanol (AR grade) Lot no. Y10B52	J.T. Baker Inc., USA
Acetonitrile (AR grade) Lot no. X51B00	J.T. Baker Inc., USA
Phosphoric acid (AR grade) Lot no. X15823	J.T. Baker Inc., USA
Sterile Water for Injection. Lot no. 41-717-XE	General Hospital Product Public Co.,Ltd. Thailand
Sodium heparin injection (5000IU/ml)	Leo Pharmaceutical, Denmark
Orange color	Hong Huat Co.,Ltd. Thailand

Drugs, reagents, and chemical substances	Supplier
Tragacanth AT50 Lot no PT76850 L00/352 no 118 OF 270	Westmayne Industrial Park, Bramston Way laindon.
Sugar for preparing syrup USP	Thaipermpoon Industry Co.,Ltd. Thailand
Sodium carboxymethylcellulose (High viscosity) 1500 CPS , BP1973, Mfg date 24/06/03, Exp date 24/01/04, Lot&Control no. 54.47412A037/2002 SA311	Srichand United Dispensary Co.,Ltd. Thailand
Methyl paraben USP XXII, Mfg date 24/07/03, Exp date 9/04/06, Lot&Control no. CD1011	Srichand United Dispensary Co.,Ltd. Thailand
Propyl paraben NFXVIII, Lot&Control no. LI2011	Srichand United Dispensary Co.,Ltd. Thailand
Sala Cider (odering agent) SDF998 Batch no. W483505	Best odour Co.,Ltd. Thailand

Instruments

Instruments	Supplier
HPLC system	
- Solvent delivery module LC-10AD	
- A model LC-10 pump	Shimadzu Corporation, Japan
- A model SIL-10A automatic sample injector	Shimadzu Corporation, Japan
- A model SPD-10AV UV spectrophotometric detector	Shimadzu Corporation, Japan
- Computer integrator, SPD-10A version 1.3 software LC-10 program	Shimadzu Corporation, Japan
- μBondapak^{TM}column (reversed-phase column C_{18}, 10μm particle size, 300 mm length, 3.9 mm internal diameter)	Waters Corporation, USA
- Insert 100 μl for 1.5 ml vials	Para-Winsor

Instruments	Supplier
Scientific refrigerator (model number UC 2021 XL-V31)	Kelvinator Scientific, Inc Conway AR, USA
Vortex mixer (Vortex-Genie™)	Scientific Industries, Inc., USA
Centrifuge machine (Hettich Universal 30F)	Dupont, Germany
Ultrasonicator (Cavitator®)	Mettler Electronic® Corp., USA
Electric analytical balance (OHAUS®)	OHAUS Corporation, USA
Electric analytical balance (Mettler AE 160)	USA
Micropipette (20-200µL)	Eppendorf® ,Germany
Micropipette (200-1000µL)	
Pipette tips (20-200µL)	
Pipette tips (200-1000µL)	
Polypropylene microtube 1.5 mL	Treff AG, Switzerland
Cellulose acetate membrane filter 0.2 µm, 47 mm diameter	Domnick Hunter
Disposable needle no.21	Terumo Corporation, Japan
Syringe insulin 100u, 1 mL	Terumo (Philippines) Corporation. Binan, Laguna, Philippines.
Syringe TB	
Parafilm (M Parafilm®)	American National Can™, USA
Aluminum foil (Diamond® Foil)	Reynold Metals Company, USA
Volumetric flask (25 mL, 50 mL, 100mL)	Pyrex® Labware, Germany
Beaker (10mL, 50mL, 100mL, 250mL, 1000mL)	
Cylinder	

Collection of plasma sample.

After the blood samples from each dose had drawn, it was immediately transferred to Eppendorf tubes containing 20 μL of 5000IU/mL sodium heparin. Then it was inverted gently and centrifuge at 5000 rpm for 10 minutes at room temperature. The plasma samples were separated and freezed at –60°C until assay. Concentration of plasma ibuprofen were determined by a technique modified from Shah et al (78).

Chromatographic condition

Column :	μ Bondapak™ column (reversed-phase column C_{18}, 10μm particle size,300 mm length, 3.9 mm internal diameter)
Insert 100μL :	Bara-Winsor
Mobile phase :	Acetonitrile: SWFI : Methanol : Phosphoric acid 40 : 55 : 5 : 0.05, v/v
Flow rate :	1.0-1.4 ml/min
Injection volume :	50-100 μL
Detector :	UV Spectrophotometric detector (wavelength 220 nm)
HPLC system :	Shimadzu solvent delivery module LC-10AD, A model LC-10AD pump, A model SIL-10A automatic sample injector, A model SPD-10AV UV spectrophotometric detector
Computer integrator :	SPD-10A version 1.3 software LC-10 program
Temperature :	Room temperature (25°C)

Preparation of mobile phase

The mobile phase was prepared by mixing acetonitrile, SWFI, methanol, and phosphoric acid at a volume ratio of 40 : 55 : 5 : 0.05, v/v respectively. It was freshly prepared and filtered through a 0.20 μm nylon membrane. The solution was deaerated 15 minutes by ultrasonicator just prior to use.

Preparation of internal standard solution

Mefenamic acid was used as internal standard for quantitative analysis in this study. It was prepared by dissolving 0.004g mefenamic acid powder in acetonitrile and adjusted volume to 100 ml with 100ml volumetric flask to yield the final concentration 40 μg/ml.

Preparation of plasma sample

1. Thaw frozen heparinized plasma at room temperature then mixing by vortex mixer for 15 seconds.

2. Transfer 100 μl of heparinized plasma into 0.2 ml eppendorf tube and add 200 μl internal standard solution containing 40 μg/ml mefenamic acid.

3. Mix for 10 seconds by vortex mixer and centrifuge at 11000 rpm for 2 minutes to pellet the precipitated proteins.

4.Transfer 100μl of the supernatant into two 1.5 ml propylene microtubes filled with 100 μl insert and inject onto the column.

Standard curve quantitation

Five concentrations of ibuprofen stock solution were prepared by weighing 0.0020, 0.0060, 0.0125, 0.0250,and 0.0500 mg of standard ibuprofen powder in 10-ml volumetric flasks and the volumes were adjusted to 10 ml with methanol in each volumetric flask. Consequently, the final concentration in each volumetric flask is 4, 12, 25, 50, and 100 μg/ml, respectively. These five concentrations were used as working standard solutions.

Five concentrations of ibuprofen in plasma were prepared by adding 20 μl of each working standard solution in five 1.5-ml eppendorf tube filled with 980 μl drug-free plasma (FFP). Then they were mixed by vortex mixer for 15 seconds. Next, transfer 400 μl of each tube into another five 1.5-ml eppendorf tube that added 800 μl internal standard solution containing 40 μg/ml mefenamic acid. They were mixed for 10 seconds by vortex mixer and centrifuged at 11000 rpm for 2 minutes. Finally, transfer 500 μl of each supernatant into 1.5 ml propylene microtube; ready to analyze by HPLC.

Standard calibration plot was constructed by least-square linear regression of the peak AUC ratio of ibuprofen to mefenamic acid versus the known corresponding ibuprofen concentrations. The unknown concentrations of ibuprofen in plasma samples were calculated from this calibration curve by inverse prediction.

Validation of HPLC method (79)

1.Separation and specificity

They were assessed by injecting 50 µl of the following samples: drug-free plasma, drug-free plasma spiked with ibuprofen concentrations of 4 µg/ml and 50 µg/ml, drug-free plasma spiked with mefenamic acid 40 µg/ml in acetonitrile, and mixture of ibuprofen 50 µg/ml and mefenamic acid 40 µg/ml into HPLC. This was to ensure that ibuprofen and mefenamic acid peaks were completely separated and were not interfered by other content from endogenous plasma constituents in term of separation and retention time.

2. Precision and accuracy

Precision of the analytical method was evaluated by intra-day and inter-day precision as following:

For intra-day precision, six replicates of ibuprofen in plasma at concentrations of 5, 20, 40, and 75µg/ml were prepared as previously described. These samples were analyzed within 1 day . For inter-day precision assessment, another six replicates of the same concentrations were analyzed in 5 different days. The concentration found in each concentration was reported as mean±SD. The precision of the assay method was evaluated as percentage of coefficient of variation (%CV) as following:

$$\%CV = \frac{SD \times 100}{\bar{x}}$$

where : SD = standard deviation

$\bar{x}$ = mean value of analyzed drug concentration

The plasma samples with the same concentrations were also prepared and kept frozen at -20^0C and were ramdomly picked up to analyze concurrently with the unknown plasma samples to control assay precision on each assay day. The accuracy

of assay procedure was evaluated by % of inaccuracy (the percentage of deviation of measured concentration from target concentration, %bias) as the following:

$$\% \text{ Bias} = \frac{\left| \text{Found concentration} - \text{Target concentration} \right| \times 100}{\text{Target concentration}}$$

The acceptable precision and accuracy were %CV < 15 and %Bias < 15.

3. Stability of ibuprofen in plasma

Six replicates of three concentration of ibuprofen in plasma (15, 30, 60 μg/ml) were prepared and analyzed immediately after preparation, then kept frozen at –60°C. These samples were assessed each week for 4 consecutive weeks. The stability was evaluated as percentage of ibuprofen concentration remaining at each time in relative to the concentration after fresh preparation.

Pharmacokinetic analysis

Plasma drug concentration-time data from all subjects were plotted. The maximum plasma concentration (C_{max}) and the time to reach maximum plasma concentration (t_{max}) were obtained from observed data of the plasma concentration-time profiles. These plasma ibuprofen concentration-time profiles from all subjects were fitted to both compartmental and noncompartmental model by assuming extravascular one compartment model with first-order absorption and first-order elimination using the WinNonlin (standard version 2.0) pharmacokinetic program. The equation of one compartment model is shown below:

$$C(t) = \frac{FDk_a \left[e^{-k_e t} - e^{-k_a t} \right]}{V(k_a - k_e)}$$

Where C(t) is plasma drug concentration, F is absolute bioavailability, k_a is absorption rate constant and k_e is elimination rate constant. V is apparent volume of distribution and D is dose, t is clock time.

Following the fitting, pharmacokinetic parameters of ibuprofen of each subject were estimated. These parameters were first-order absorption rate constant (k_a), first-

order elimination rate constant (k_e), elimination half-life ($t_{1/2}$), apparent volume of distribution (Vd/F), area under the plasma concentration versus time curve from time zero to infinity ($AUC_{0-\infty}$) and also apparent total body clearance (Cl/F).

The noncompartmental analysis was also conducted to determine the area under the plasma concentration versus time curve from time zero to the last time that plasma level still above the limit of quantitation, $AUC_{0-tlast}$; the area under the plasma concentration versus time curve from time zero to infinity, $AUC_{0-\infty}$; apparent volume of distribution, Vd/F; and apparent total body clearance, Cl/F. The $AUC_{0-tlast}$ was estimates by the WinNonlin program using linear trapezoidal method. The area under the curve extrapolated to infinity, $AUC_{0-\infty,}$ was obtained by adding the remaining area under the curve from t_{last} to infinity as shown in the following equation:

$$AUC_{0-\infty} = AUC_{0-tlast} + AUC_{tlast-\infty}$$

$$AUC_{tlast-\infty} = C_{last} / \lambda_z$$

This remaining area was calculated by dividing the last detectable concentration by the terminal elimination rate constant (λ_z) which is estimated as the slope of terminal natural log-linear portion of plasma concentration time course.

Statistics

1. Population characteristic

1.1 Continuous variables such as gestational age, birth weight, apgar score, age of drug administration, and daily fluid intake in the first week in each groups were summarized with descriptive statistics expressed as mean±SD. The mean difference in each continuous variables between the two groups were tested with independent-sample t-test.

1.2 Categorial variables such as sex, number of silent PDA subjects, and antenatal steroid were analyzed with the percentage in each variables and the mean difference in each variables between the two groups were compared by chi-square or Fisher's Exact test when the expected count less than 5 were presented more than 20%

2. Efficacy data

2.1 The proportion of successful PDA closure and the reduction of silent PDA after drug administration course in each groups were compared with Fisher's Exact test.

2.2 The distribution of PDA according to the birthweight or gestational age between the two groups were tested with Chi-Square or Fisher's Exact test.

3. Safety data

3.1 Continuous variables such as the number of mechanical ventilation days, oxygen therapy days, start feeding days, full feeding days, and length of stay were summarized with descriptive statistics by mean±SD. The mean comparison between the two groups were analyzed with independent sample t-test.

3.2 Categorial variables such as the proportion of bleeding disorder, BPD, abdominal distention, feeding difficulty, NEC, IVH, ROP, and survival rate between two groups were analyzed with chi-square.

3.3 The renal laboratory values (BUN and creatinine) and the hematological laboratory values (INR, PTT, and platelet count) between the groups in each days were compared with independent-sample t-test. The mean comparison of each values before and after drug administration were analyzed by paired-sample t-test.

4. Pharmacokinetics data

4.1 The pharmacokinetic parameters were analyzed by descriptive statistics expressed as mean±SD if they were normal distribution or median mean±IQR if they showed right skewness. The extravascular one compartment model was assumed as the following formula;

$$C_p = \frac{F \ Dose \ k_a \ t \ (e^{-ket} - e^{-kat})}{V_d \ (k_a - k_e)}$$

While

C_p = plasma concentration

F = absolute bioavailability

k_a = absorption rate constant

k_e = elimination rate constant

t = time

V_d = volume of distribution

When pharmacokinetic parameters from each subjects were obtained, the standard two stage (STS) method were used to predicted population pharmacokinetic. Drug concentration data after the first dose administration were used to predict drug concentration after the second and the third doses were taken.

4.2 The mean difference between the observed and the predicted concentrations from the pooling of all subjects in each time point were tested with independent-sample t-test.

4.3 The mean difference in pharmacokinetic parameters categorized by sex, gestational age, and birthweight in each groups were tested with independent-sample t-test.

CHAPTER 4

RESULT

Clinical characteristics between the groups.

There were one hundred and thirteen patients admitted during this 10-month-period. Seventy-one cases (62.83%) were not recruited in the study because of unstable clinical conditions thus making 42 cases recruited in the study. These 42 cases were randomized to the ibuprofen (n = 22) and control group (n = 20). There were no significant differences in sex, gestational age, birth weight, Apgar scores at 1 and 5 minutes, antenatal steroid, age of drug administration, and the presence of PDA by echocardiogram between the groups before enrollment. The data were shown in the Table45. The daily fluid intake was not differed significantly between the groups as shown in the table 46.

Table 45 Demographic data of the recruited subjects in both study groups.

Clinical characteristics	Ibuprofen group (n=22)	Control group (n=20)	p-value
Sex (M:F) (%)	14 : 8 (1.8 : 1)	13 : 7 (1.86 : 1)	0.927
Gestational age (weeks)	30.64±1.76	30.20±2.14	0.473
Birth weight (gram)	1279.64±80.33	1214.50±217.52	0.295
Apgar score			
1 min	6.38±2.52	7.10±1.71	0.291
5 min	7.90±2.17	8.15±1.53	0.679
Antenatal steroid (%)	13/22 (59.09%)	10/20 (50.00%)	0.554
Age of drug administration (hrs)	23.93±9.46	22.88±10.13	0.732
Silent PDA	14 (70.00%)	15 (68.18%)	0.899

Table 46 Daily fluid intake data during the first week in both study groups.

Day	Fluid intake (ml/kg/day)		P – value
	Ibuprofen group	Control group	
1	102.10±21.35	98.95±26.86	0.674
2	115.94±20.85	111.90±25.85	0.578
3	129.80±36.88	127.95±26.59	0.855
4	146.84±24.52	145.54±32.75	0.884
5	156.61±31.22	154.38±48.49	0.862
6	173.33±30.91	162.21±44.63	0.357
7	186.83±34.45	172.57±39.60	0.237

Incidence of PDA in relation to birthweight and gestational age

Birthweight and gestational age were important factors for PDA closure. We divided subjects into two groups for each factors. Table 47 demonstrated the incidence of PDA in relation to birthweight and gestational age. The incidence of PDA on day 0 was higher in the group with lower birthweight and gestational age without statistical significance.

Table 47 Distribution of PDA during study period by birthweight and gestational age.

Factors		PDA incidence	Percentage	p-value
Birthweight	< 1200 g	12/17	70.6	0.859
	≥ 1200 g	17/25	68.0	
Gestational age	< 30 wks	12/14	85.7	0.159
	≥ 30 wks	17/28	60.7	

PDA evaluation by echocardiogram

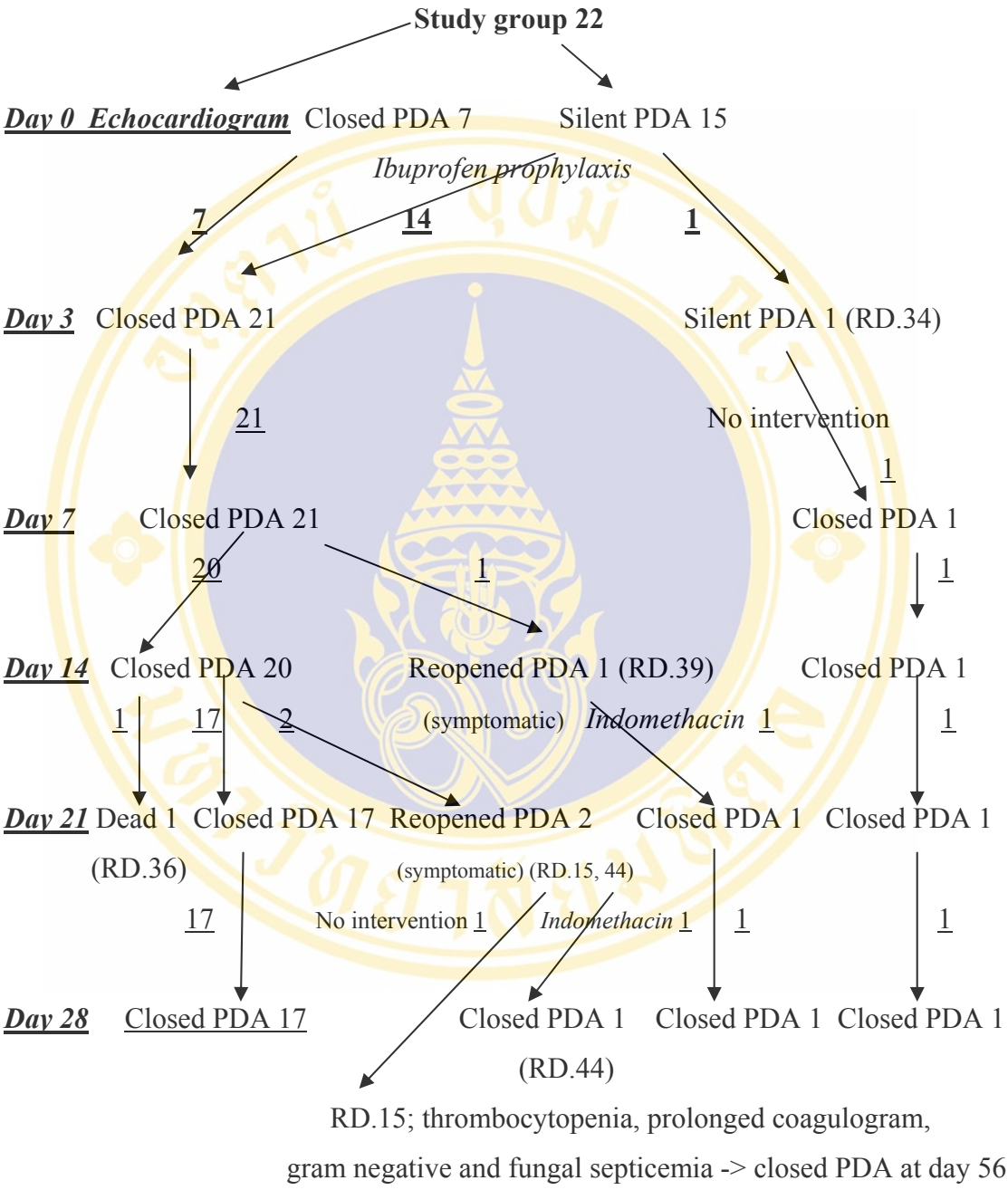

Figure 9 Diagram of summarizing echocardiogram result during study period in the study group.

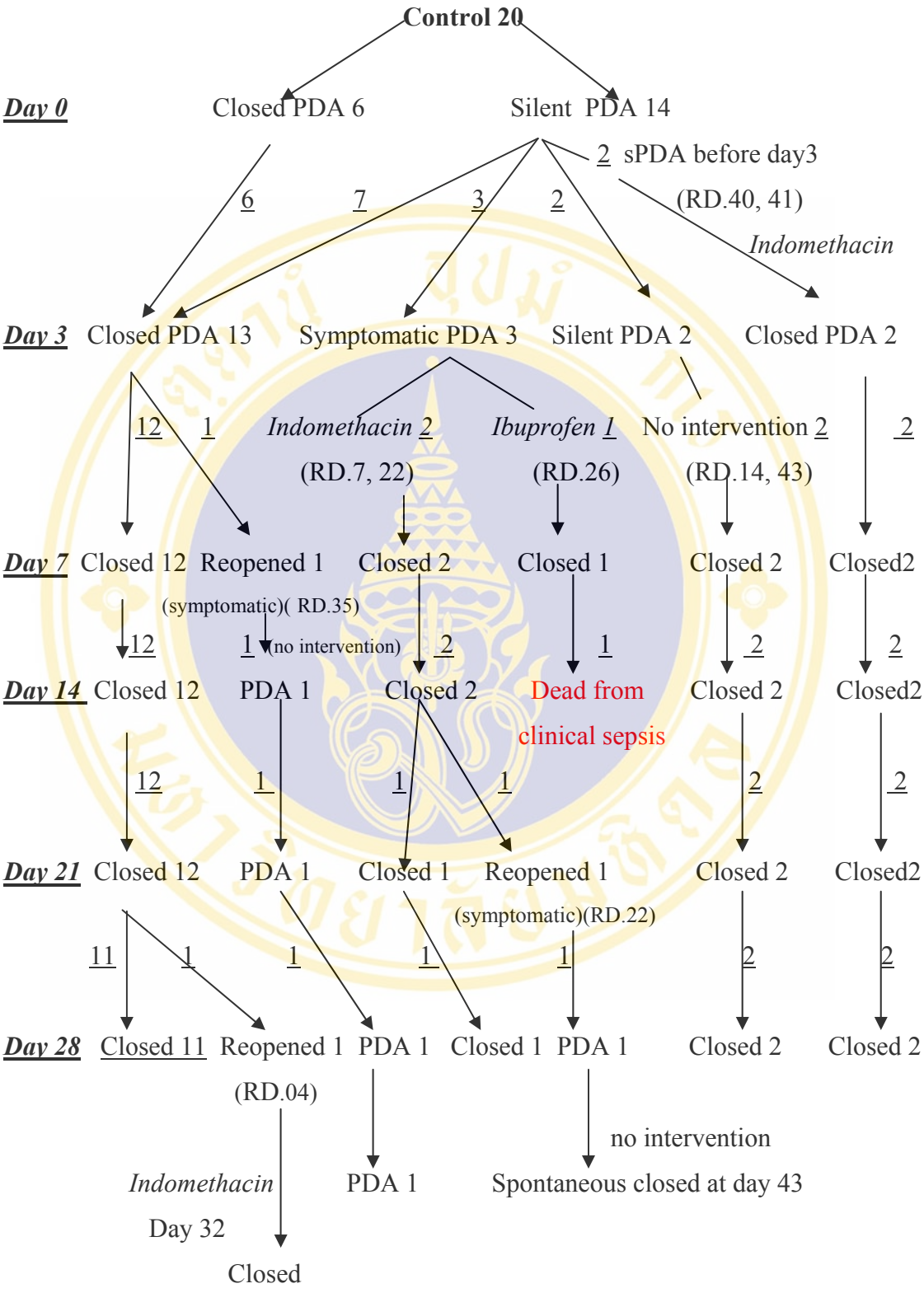

Figure 10 Diagram of summarizing echocardiogram result during study period in the control group.

Study group

1. Echocardiogram on day 0 of the twenty two patients in the study group revealed asymptomatic PDA in 15 cases before the ibuprofen administration.

2. Echocardiogram on day 3 revealed only one patient with PDA without symptoms (randomized no.34). The efficacy of oral ibuprofen was 95.45% by this day.

3. Echocardiogram on day 7 revealed closed ductus in all 22 cases. The efficacy of oral ibuprofen was 100% by this day.

4. Echocardiogram on day14 revealed reopen, symptomatic PDA in one case and he was treated with indomethacin (randomized no.39).

5. One of the patients in this group (randomized no 36) died from staphylococcus coagulase negative septicemia on day 16. Echocardiogram on day 21 of the remaining 21 cases revealed two more (randomized no 15 and 44) with reopen, symptomatic PDA. Only one case (randomized no 44) received one course of indomethacin because the other one (randomized no 15) had coagulopathy from fungal septicemia.

5. Echocardiogram on day 28 revealed closed ductus arteriosus in 20 cases with the only remaining one (randomized no 15) had PDA. This patient had been followed and found to have ductal closure spontaneously on day 56.

Control group

1. Echocardiogram on day 0 of the twenty patients in the control group revealed asymptomatic PDA in 14 cases before the placebo administration.

2. Echocardiogram on day 3 showed that all six cases with previously closed ductus arteriosus had no reopening. Two cases (randomized no 40 and 41) had symptomatic PDA before day 3 and had been treated with indomethacin on day 2 and day 3 respectively. Three cases (randomized no.7, 22 and 26) had symptomatic PDA on day 3 and had been treated with indomethacin in two (randomized no.7 and 22) while the other one (randomized no 26) treated with ibuprofen.

3. Echocardiogram on day 7 revealed that three cases of reopening PDA had no residual ductus after treatment. One of the cases with previously closed ductus on day 3 (randomized no.35) had reopening without specific treatment because of thrombocytopenia from clinical sepsis. The ductus of this latter case closed later.

4. One case (randomized no. 26) died from clinical sepsis at day 11. Echocardiogram on day 14 of the other 18 cases revealed no PDA.

5. Echocardiogram on day 21 showed that one case (randomized no.22) had reopening PDA without specific treatment because of thrombocytopenia from clinical sepsis. This case had been treated for symptomatic PDA on day 3 with indomethacin. This patient had been followed and found to have ductal closure spontaneously on day 43. The case with reopen ductus on day 7 (randomized no.35) still had PDA. Echocardiogram of the other 17 cases revealed no PDA.

6. Echocardiogram on day 28 showed that one more case (randomized no.4) had reopening PDA and been treated with indomethacin. The case with reopen ductus on day 7 (randomized no.35) still had PDA. Echocardiogram of the other 16 cases revealed no PDA.

Primary outcome

Table 48 Primary outcome at 3 days after drug administration.

PDA status	Ibuprofen group	Control group	P – value
Closed within 72 hours	21/22 (95.45%)	13/20 (65.00%)	0.018
-No reopening throughout the study period	*17/21 (80.95%)*	*11/13 (84.62%)*	
-Reopening during study period	*3/21 (14.29%)*$^{\Delta}$	*2/13 (15.38%)*$^{\Delta\Delta}$	
-Death during study period	*1/21 (4.76%)*		

$^{\Delta}$ Reopening at day 21, day 14, and day 20

$^{\Delta\Delta}$ Reopening at day 28 and day 7-28

At day 3, the percentage of PDA closure in ibuprofen group was significantly higher than control group (95.45% vs 65.00%; $p<0.05$). Back up treatment of symptomatic PDA with indomethacin was success in two cases of ibuprofen group (50.00%) and six cases of control group (66.67%).

Prevention of silent PDA

Table 49 Silent PDA reduction evaluated at day3 after drug administration.

Number of subjects	Ibuprofen group	Control group	p-value
Day 0; silent PDA	15	14	
Day3;unclosed PDA	1	7	
Silent PDA reduction	14 (93.3%)	7 (50.0%)	0.014

There were fifteen subjects in ibuprofen group and fourteen subjects in control group presented silent PDA proven by echocardiogram before taking intervention. According to the table 49, the incidence of silent PDA reduction was higher in ibuprofen group than in control group with statistically significance (93.3% vs 50.0%; $p < 0.05$).

Assessment of other outcome variables

Many previous studies reported that ibuprofen affect the major organ less commonly than indomethacin. We evaluated the side effects of ibuprofen to the major organs as following.

Table 50 Secondary outcome variable assessment

Outcomes	Ibuprofen group	Control group	p-value
Bleeding disorder	13/21 (61.9%)	10/20 (50.0%)	0.443
Respiratory tract			
-PPHN	0	0	
-BPD	7/21 (33.3%)	2/20 (10.0%)	0.130
-Days of mechanical ventilation	11.66	7.59	0.332
-Days of oxygen therapy	15.27	9.98	0.199
GI tract			
-Abdominal distention	10/22 (45.5%)	6/20 (30.0%)	0.303
-Feeding difficulty	12/21 (57.1%)	8/20 (40.0%)	0.272
-NEC	8/21 (38.1%)	6/20 (30.0%)	0.585
-Days of start feeding	6.23	3.70	0.029*
-Days of full feeding	29.71	26.94	0.677
IVH	2/21 (9.5%)	4/19 (21.1%)	0.398
ROP	3/21 (14.3%)	2/19 (10.5%)	1.000
Length of hospital stay	61.90	53.65	0.339
Survival rate	21/22 (95.45%)	19/20 (95.00%)	1.000

Table 51 Laboratory results

Variables	Ibuprofen group	Control group	p-value
BUN			
Day 1	12.22 ± 4.35	13.35 ± 4.15	0.334
Day 3	15.14 ± 6.50	17.84 ± 13.65	0.422
p-value	0.102	0.174	
Creatinine			
Day 1	0.82 ± 0.18	0.91 ± 0.22	0.211
Day 3	0.87 ± 0.17	0.86 ± 0.22	0.883
p-value	0.285	0.470	
INR			
Day 1	1.43 ± 0.17	1.41 ± 0.21	0.598
Day 3	1.28 ± 0.15	1.69 ± 1.78	0.300
p-value	0.017	0.513	
PTT			
Day 1	61.62 ± 15.88	56.79 ± 18.30	0.381
Day 3	56.42 ± 13.81	49.06 ± 16.57	0.139
p-value	0.229	0.040	
Platelet			
Day 1	224,545.45 ± 64,680.70	219,400 ± 50,988.50	0.778
Day 3	185,590.91 ± 55,825.12	184,368 ± 84,343.40	0.957
p-value	0.005	0.103	

Renal system

BUN and Cr levels before and after drug administration did not differ significantly nor did the levels between the study and control group.

Hematological system

International normalized ratio (INR) and platelet count at day 3 decreased significantly from day 0 but these values were within normal range and did not differ significantly from the control group.

Gastrointestinal system

Despite these normal coagulogram, there were thirteen cases with upper gastrointestinal bleeding. The time needed to start feeding was significantly longer in the study group than in the control group. Although there was no statistical significance, the incidence of abdominal distention, feeding difficulty, NEC and time needed to reach full feeding tended to be higher in the ibuprofen than in the control group.

Respiratory system

There was no persistent pulmonary hypertension of the newborn (PPHN) in this study. There were trends toward the higher incidence of BPD, longer time of mechanical ventilation and oxygen therapy in the ibuprofen than in the control group, although the differences were not statistically significant.

Other variables

There were no statistical differences between both groups in the incidence of IVH, retinopathy of prematurity (ROP) and the duration of hospital stay. The incidence of IVH tended to be higher in the control than in the study group, ROP tended to be higher in the study than in the control group and patients in the ibuprofen group tended to have longer duration of hospitalization than in the control group.

Drug concentration data of all subjects in the study group

During the study period, we adjusted the drug sampling time into three intervals according to the concentration-time curve of each period.

First interval

The drug sampling times were 15 mins, 30 mins, 45 mins, 1 hr, 2 hrs, 4 hrs, and 6 hrs after the first dose was given. Before 30 mins and after 1 hr of giving the second and the third dose, drug sampling were also taken. There were two subjects (randomized no. 2 and no.3) that followed this protocol. Drug concentration data and graph were shown in table 52 and figure… respectively.

Table 52 Drug concentration data of the two subjects following the first sampling time interval.

Time	Concentration (mcg/ml) of subject number	
	1; RD02	2; RD03
0 min	Dose 1	Dose 1
15 mins	1.93	1.09
30 mins	2.82	
45 mins		1.18
1 hr	4.26	
2 hrs	9.33	2.60
4 hrs	30.78	5.20
6 hrs	32.49	5.56
23.5 hrs	36.23	
24 hrs	Dose 2	
25 hrs	34.48	
26 hrs		15.49
26.5 hrs		Dose 2♠
27.5 hrs		15.14
47.5 hrs	33.09	
48 hrs	Dose 3	
49 hrs	42.77	
50 hrs		25.97
50.5 hrs		Dose 3
51.5 hrs		23.05

** The second dose of subject randomized no.3 was postponed for 2.5 hours because he/she had oliguria (urine output less than 0.5 ml/kg/hr) with shock. He/She was treated by PRC and fluid replacement until stable clinical condition. The third dose was given apart from second dose 24 hours.

According to the figure…..and …. , the concentration-time curve of two subjects couldn't predict maximum concentration and elimination pattern obviously in the first six hours so these data couldn't be fitted to Win Nonlin program for pharmacokinetic parameters calculation. Consequently, we reduced the sampling points during the first hour but distributed the sampling points during six and twenty-four hours instead.

Next interval

The new drug sampling times after giving first dose were 1, 2, 4, 5, 7, 10, and 18 hrs. For the second and the third dose, drug sampling were taken before 30 mins and after 8 hrs of drug administrations. There were three subjects followed this new protocol and their drug concentration patterns were shown in table 53 and figure…….

Table 53 Drug concentration data of the three subjects following the second sampling time interval.

Time (hrs)	Concentration (mcg/ml) of subject number		
	3; RD06	**4; RD08**	**5; RD10**
0	**Dose 1**	**Dose 1**	**Dose 1**
1	2.05	20.08	6.08
2	3.28	25.78	9.28
4	6.71	26.61	13.72
5	22.00	26.94	14.64
7	23.51	24.97	16.94
10	26.44	23.58	19.36
18	20.22	16.00	17.25
23.5	20.22	11.83	13.64
24	**Dose 2**	**Dose 2**	**Omitted dose 2**
32	37.84	23.78	8.22
47.5	32.84	6.08	2.67
48	**Dose 3**	**Omitted dose 3**	**Omitted dose 3**
56	47.35	3.11	1.75

**Subject no.4: The third dose was omitted because of coagulopathy together with UGIB. FFP and IV Ranitidine were given in order to correct these abnormal conditions. However, sampling time after the second dose was continued as the schedule.

***Subject no.5. The second and the third doses couldn't be taken since the coffee ground content and abnormal coagulogram were found after the first dose. He/She was treated by FFP and IV Ranitidine; nevertheless, his/her sampling time after the first dose was still followed as setting protocol.

From the concentration-time curves of three subjects, we could see absorption and elimination pattern more clearly than the first protocol. Although this protocol was better than the previous protocol, the sampling points between ten and eighteen hours after the first dose was less than the time between one and ten hours. Therefore, we reduced the sampling time points before ten hours but extended the sampling time points after ten hours for the last protocol.

Last interval

The time of drug sampling were 2, 4, 8, 10, 12, 14, and 18 hours after first dose. The sampling time for the second and the third doses were 30 min before and 8 hours after each dose. There were seventeen subjects followed this schedule and their plasma drug concentration data and graphs were shown in table 54 and figure…..,respectively.

Table 54. Drug concentration data of the seventeen subjects following the third sampling time interval.

Time (hrs)	Concentration (mcg/ml) of subject number								
	6; RD12	7; RD13	8; RD15	9; RD18	10; RD19	11; RD21	12; RD24	13; RD25	14; RD27
0	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**
2	6.93	3.07	7.05	29.36	30.97	35.5	16.71	4.98	35.09
4	13.74	5.99	7.85	25.39	27.27	43.41	24.85	7.07	35.53
8	16.98	8.64	8.89	34.02	33.73	38.67	34.86	16.77	25.29
10	10.35	8.34	16.61	35.94	26.17	48.24	36.78	17.82	26.72
12	15.97	11.95	15.28	36.72	33.49	35.95	40.46	19.21	34.03
14	17.47	11.58	19.85	35.39	33.28	28.09	38.11	19.03	33.19
18	14.77	12.15	15.86	34.92	28.78	39.74	35.64	31.64	30.01
23.5	16.32	12.5	19.51	33.03	27.31	45.86	39.16	30.94	29.14
24	**Dose 2**		**Dose 2**					**Dose 2**	
32	61.09	13.07	35.39	57.44	43.35	44.48	24.8	52.67	62.72
39.5		12.98							
40		**Dose 2**							
47.5	45.9		34.15	49.21	39.52	46.39	27.1	47.26	50.38
48	**Dose 3**	43.88	**Dose 3**					**Dose 3**	
56	85.84		48.49	47.81	67.49	57.09	27.96	42.05	
63.5		30.17							
64		**Dose 3**							
72		66.5							
73							15.85		
73.5							**Dose 2**		
81.5							68.83		
97							32.62		
97.5							**Dose 3**		**Omitted dose3**
105.5							100.79		

**Subject no.7. The second dose was postponed for 16 hours because of prolonged PTT and UGIB. FFP and Ranitidine inj were given for correcting these abnormal conditions respectively. The third dose was given apart from the second dose 24 hours.

**Subject no.12. The second dose was 49.5 delayed according to prolonged PTT and much coffee ground content. He/She was corrected by FFP, Ranitidine inj, and gastric lavage. The third dose schedule didn't change.

**Subject no. 14. The third dose was omitted because the subject's condition was unstable from clinical sepsis and hyperbilirubinemia. The exchange transfusion 23 cycles were given.

Table 54 Drug concentration data of the seventeen subjects following the third sampling time interval (continued).

Time (hrs)	Concentration (mcg/ml) of subject number							
	15; RD30	**16; RD32**	**17; RD34**	**18; RD36**	**19; RD38**	**20; RD39**	**21; RD42**	**22; RD44**
0	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**	**Dose 1**
2	5.14	16.63	45.86	6.78	12.3	10.8	12.94	40.62
4	34.2	54.99	33.01	9.26	43.42	15.94	15.35	47.17
8	35.24	34.1	21.22	13.78	27.69	61.23	15.03	48.31
10	26.5	27.87	18.62	15.48	28.41	58.39	14.75	41.51
12	30.78	24.24	15.23	15.43	31.72	52.06	15.67	46.81
14	26.02	18.28	14.93	15.86	33.93	52.97	15.18	39.89
18	33.79	15	10.57	14.64	42.54	40.78	18.46	52.83
23.5	32.03	9.85	7.37	14.08	33.84	44.15	12.48	30.58
24	**Dose 2**							
32	59.78	17.01	21.57	27.54	42.2	71.92	60.56	49.72
47.5	46.55	5.02	10.73	27.99	51.58	72.68	33.87	47.46
48	**Dose 3**				**Omitted dose 3**	**Dose 3**		
56	61.34	26.35	25.04	53.71		129.54	65.69	62.63

**Subject no.19. The third dose couldn't be given since coffee ground content and prolonged PTT were found before drug administration. Ranitidine inj and vitamin K 2 mg were given for correcting bleeding disorder.

Drug level data for 22 subjects from three interval were analyzed by Win Nonlin program; however, five subjects couldn't be selected to analyze by this program. The first sampling schedule data of the two subjects couldn't predict C_{max} and ke that essential for running program. Additionally, data of the last three subjects from the third sampling schedule couldn't predict k_e because the elimination pattern couldn't be seen obviously during the first twenty-four hours. In summary, only seventeen subjects were selected for predicting drug concentration at each time and essential pharmacokinetic parameters by Win Nonlin program as shown in table 55.

Table 55 Observed and Predicted concentration in each time points of the seventeen subjects

Time (hrs)	Drug concentration (mcg/ml) of selected subjects by Win Nonlin program					
	sbj 3; RD06		sbj 4; RD08		sbj 5; RD10	
	Observed	Predicted	Observed	Predicted	Observed	Predicted
0	0.00	0.00	0.00	0.00	0.00	0.00
1	2.05	4.51	20.08	19.17	6.08	4.45
2	3.28	8.34	25.78	26.26	9.28	8.12
4	6.71	14.28	26.61	28.00	13.72	13.50
5	22.00	16.53	26.94	26.92	14.64	15.38
7	23.51	19.90	24.97	24.05	16.94	17.91
10	26.44	22.80	23.58	19.88	19.36	19.41
18	20.22	23.49	16.00	11.85	17.25	16.79
23.5	20.22	21.42	11.83	8.30	13.64	13.28
32	37.84	38.46	23.78	27.38	8.22	8.38
47.5	32.84	32.33	6.08	10.05	2.67	3.10
56	47.35	46.79			1.75	1.72

Table 55 Observed and Predicted concentration in each time points of the seventeen subjects (continued).

Time (hrs)	Drug concentration (mcg/ml) of selected subjects by Win Nonlin program					
	sbj 6; RD12		sbj 8; RD15		sbj 9; RD18	
	Observed	Predicted	Observed	Predicted	Observed	Predicted
0	0.00	0.00	0.00	0.00	0.00	0.00
2	6.93	4.72	7.05	4.81	29.36	13.79
4	13.74	8.74	7.85	8.54	25.39	23.54
8	16.98	15.09	8.89	13.60	34.02	34.33
10	10.35	17.59	16.61	15.27	35.94	36.65
12	15.97	19.71	15.28	16.51	36.72	37.55
14	17.47	21.52	19.85	17.42	35.39	37.41
18	14.77	24.39	15.86	18.52	34.92	35.07
23.5	16.32	27.09	19.51	19.02	33.03	29.66
32	61.09	44.61	35.39	32.25	57.44	54.97
47.5	45.90	58.38	34.15	35.91	49.21	38.66
56	85.84	76.25	48.49	48.09	47.81	60.40

Table 55 Observed and Predicted concentration in each time points of the seventeen subjects (continued).

Time (hrs)	Drug concentration (mcg/ml) of selected subjects by Win Nonlin program					
	sbj 10; RD19		sbj 11; RD21		sbj 12; RD24	
	Observed	Predicted	Observed	Predicted	Observed	Predicted
0	0.00	0.00	0.00	0.00	0.00	0.00
2	30.97	30.21	35.50	35.72	16.71	19.07
4	27.27	31.61	43.41	40.72	24.85	23.92
8	33.73	30.01	38.67	38.10	34.86	25.34
10	26.17	29.15	48.24	36.17	36.78	25.34
12	33.49	28.32	35.95	34.30	40.46	25.28
14	33.28	27.51	28.09	32.53	38.11	25.20
18	28.78	25.96	39.74	29.24	35.64	25.03
23.5	27.31	23.97	45.86	25.25	39.16	24.80
32	43.35	51.20	44.48	58.23	24.80	49.79
47.5	39.52	40.89	46.39	38.57	27.10	48.62
56	67.49	66.15	57.09	68.85	27.96	47.93
73					15.85	46.58
81.5					68.83	45.92
97					32.62	44.74
105.5					100.79	69.45

Table 55 Observed and Predicted concentration in each time points of the seventeen subjects (continued).

Time (hrs)	Drug concentration (mcg/ml) of selected subjects by Win Nonlin program					
	sbj 13; RD25		sbj 14; RD27		sbj 15; RD30	
	Observed	Predicted	Observed	Predicted	Observed	Predicted
0	0.00	0.00	0.00	0.00	0.00	0.00
2	4.98	6.03	35.09	34.02	5.14	14.37
4	7.07	10.97	35.53	33.47	34.20	23.25
8	16.77	18.15	25.29	32.41	35.24	31.39
10	17.82	20.63	26.72	31.90	26.50	32.72
12	19.21	22.51	34.03	31.39	30.78	33.03
14	19.03	23.89	33.19	30.88	26.02	32.71
18	31.64	25.40	30.01	29.90	33.79	31.02
23.5	30.94	25.54	29.14	28.61	32.03	27.83
32	52.67	41.38	62.72	59.13	59.78	54.27
47.5	47.26	42.06	50.38	52.19	46.55	43.56
56	42.05	54.39			61.34	67.06

Table 55 Observed and Predicted concentration in each time points of the seventeen subjects (continued).

Time (hrs)	Drug concentration (mcg/ml) of selected subjects by Win Nonlin program					
	sbj 16; RD32		sbj 19; RD38		sbj 20; RD39	
	Observed	Predicted	Observed	Predicted	Observed	Predicted
0	0.00	0.00	0.00	0.00	0.00	0.00
2	16.63	28.35	12.30	19.53	10.80	19.24
4	54.99	36.11	43.42	27.61	15.94	31.70
8	34.10	30.65	27.69	31.65	61.23	44.47
10	27.87	25.49	28.41	31.66	58.39	47.29
12	24.24	20.62	31.72	31.29	52.06	48.73
14	18.28	16.40	33.93	30.74	52.97	49.27
18	15.00	10.11	42.54	29.51	40.78	48.74
23.5	9.85	5.07	33.84	27.81	44.15	46.42
32	17.01	32.37	42.20	57.00	71.92	86.41
47.5	5.02	5.31	51.58	49.22	72.68	80.69
56	26.35	32.45			129.54	117.05

Table 55 Observed and Predicted concentration in each time points of the seventeen subjects (continued).

Time (hrs)	Drug concentration (mcg/ml) of selected subjects by Win Nonlin program			
	sbj 21; RD42		sbj 22; RD44	
	Observed	Predicted	Observed	Predicted
0	0.00	0.00	0.00	0.00
2	12.94	7.78	40.62	39.53
4	15.35	13.38	47.17	46.91
8	15.03	20.17	48.31	44.11
10	14.75	22.07	41.51	41.48
12	15.67	23.31	46.81	38.91
14	15.18	24.06	39.89	36.46
18	18.46	24.59	52.83	32.01
23.5	12.48	24.11	30.58	26.76
32	60.56	42.47	49.72	64.40
47.5	33.87	42.65	47.46	39.01
56	65.69	59.13	62.63	73.69

To validate that the observed concentration was not different from the predicted concentration at each time point, all the same sampling points in each subjects were pooled in order to compare the mean difference as following table.

Table 56 Comparison of observed and predicted concentration in each time points of seventeen subjects

Times (hrs)	N	Average concentration (mcg/ml)		p-value
		Observed	Predicted	
0	17	0.00±0.00	0.00±0.00	1.0000
1	3	9.40±9.46	9.38±8.48	0.9850
2	17	17.84±12.38	18.82±11.65	0.5369
4	17	26.07±15.07	24.49±11.57	0.4356
5	3	21.19±6.19	19.61±6.36	0.5032
7	3	21.81±4.28	20.62±3.13	0.4661
8	14	30.77±13.92	29.25±9.79	0.3827
10	17	28.56±12.44	27.97±8.97	0.7054
12	14	30.89±11.60	29.39±8.84	0.3390
14	14	29.33±10.77	29.00±8.71	0.8324
18	17	28.72±11.67	25.98±9.03	0.1731
23.5	17	26.46±11.36	23.82±9.21	0.1711
32	17	44.29±17.91	47.22±17.34	0.3403
47.5	17	37.57±18.67	38.89±19.12	0.5075
56	14	55.10±29.86	58.57±25.56	0.1935

From the table 56, there were no difference (p-value > 0.05) between the average observed and predicted concentration in each time point. Consequently, data from the first dose could be used to predict the concentration after the second and the third doses were administered with nearly equal to actual concentration.

The initial pharmacokinetics parameters (V_d/F, k_a, k_e) in each subjects were calculated by Win Nonlin program and all of these initial pharmacokinetics parameter were used to calculate the secondary pharmacokinetics parameters as the following table 57.

Table 57 Clinical characteristics and/or pharmacokinetics parameters of ibuprofen suspension in seventeen subjects that could be fitted by Win Nonlin program and in three subjects that couldn't be fitted by Win Nonlin program in the study group.

	Clinical charcteristics					Primary PK parameters				Secondary PK parameters			
Subject no.	BW	GA	Echo Day 0	Echo Day 3	Conc at t 23.5	Vd/F	Ka	Ke	AUC	T1/2	CL	Tmax	Cmax
For 17 subjects that could be fitted by Win Nonlin program													
RD06	1400	31	+	-	20.22	0.3655	0.1275	0.0324	1182.80	21.4	0.0118	14.41	24.02
RD08	1400	32	-	-	11.83	0.3992	0.8367	0.0648	541.46	10.7	0.0259	3.31	28.30
RD10	1100	32	-	-	13.64	0.2437	0.1082	0.0772	584.61	8.98	0.0188	10.89	19.48
RD12	1160	29	+	-	16.32	0.3623	0.0797	0.0000	7850970.93	169959.25	0.0000	124.00	32.00
RD15	1100	29	+	-	19.51	0.4751	0.1175	0.008	2890.54	86.53	0.0038	24.52	19.03
RD18	1564	34	+	-	33.03	0.1517	0.0783	0.0796	1294.43	8.7	0.0121	12.66	37.61
RD19	1074	28	+	-	27.31	0.3223	1.2947	0.0145	2297.01	47.77	0.0047	3.51	31.67
RD21	955	28	+	-	45.86	0.2078	0.8253	0.0266	1715.21	26.01	0.0055	4.30	40.76
RD24	1260	30	+	-	39.16	0.4896	0.6784	0.0017	15319.59	412.61	0.0008	8.87	25.35
RD25	1190	29	+	-	30.94	0.1740	0.0485	0.0465	1471.82	14.91	0.0081	21.06	25.70
RD27	1040	30	-	-	29.14	0.3013	4.9571	0.0081	4284.46	86.04	0.0024	1.30	34.16
RD30	1410	31	-	-	32.03	0.3175	0.2035	0.0244	1808.20	28.42	0.0077	11.84	33.04
RD32	1280	31	-	-	9.85	0.1964	0.3471	0.128	509.08	5.42	0.0251	4.55	36.38
RD38	1430	32	-	-	33.84	0.4086	0.4164	0.0109	3209.41	63.57	0.0045	8.98	31.73
RD39	1110	30	+	-	44.15	0.1855	0.1973	0.0132	4538.03	52.56	0.0024	14.70	49.30
RD42	1520	31	+	-	12.48	0.4900	0.1466	0.0127	2440.13	54.52	0.0062	18.27	24.59
RD44	1214	32	+	-	30.58	0.2214	0.6934	0.0326	1683.42	21.28	0.0072	4.63	47.16
Distribution	Normal	Normal			Normal	Normal	Right skewness						Normal
Mean	1247	31			26.46	0.3125	0.6562	0.0342	464514.18	10053.45	0.0087	17.16	31.78
SD	179	2			11.36	0.1146	1.1646	0.0348	1903449.42	41206.89	0.0078	28.32	8.67
Med	1214	31			29.14	0.3175	0.2035	0.0244	1808.20	28.42	0.0062	10.89	31.73
Q1	1100	29			16.32	0.2078	0.1175	0.0109	1294.43	14.91	0.0038	4.55	25.35
Q3	1400	32			33.03	0.3992	0.6934	0.0465	3209.41	63.57	0.0118	14.70	36.38
IQR	300	3			16.71	0.1913	0.5759	0.0356	1914.99	48.66	0.0080	10.14	11.02
Max	1564	34			45.86	0.4900	4.9571	0.1280	7850970.93	169959.25	0.0259	124.00	49.30
Min	955	28			9.85	0.1517	0.0485	0.0000	509.08	5.42	0.0000	1.30	19.03
For 3 subjects that couldn't be fitted by Win Nonlin program													
RD13	1140	28	+	-	12.5	can't be calculated							
RD34	1510	33	+	+	7.37	can't be calculated							
RD36	1485	32	+	-	14.08	can't be calculated							

The initial parameters were V_d/F, ka, and ke. Firstly, the apparent volume of distribution (V_d/F) of ibuprofen was 0.3175 (0.2078-0.3992) l/kg. Next, the absorption rate constant (k_a) was 0.2035 (0.1175-0.6934) hr^{-1}. Lastly, the elimination rate constant was 0.0244 (0.0109-0.0465) hr^{-1}.These initial parameters were used to calculate the secondary parameters. These are $t_{1/2,}$ CL/F, AUC, C_{max}, and T_{max}. First, the elimination half-life ($t_{1/2}$) was 28.42 (19.41-63.57) hrs. Second, the apparent clearance (CL/F) was 0.0076 l/hr. Third, area under the concentration curve (AUC) was 1808.20(1294.4258-3209.4115). Next, maximum concentration (C_{max}) was 31.73

(25.35-36.38) mcg/ml. Last, time to maximum concentration (tmax) was 10.89 (4.55-14.70) hrs.

From the above predicted pharmacokinetic data, we can categorize each pharmacokinetic parameters by sex, GA, and BW as shown in the table 58.

Table 58 Effect of sex, gestational age, and birth weight on the pharmacokinetics parameters.

Factors	Initial parameters			Secondary parameters			
	Vd/F	Ka	Ke	AUC	KeT1/2	Tmax	Cmax
Sex							
Male	0.3016±0.1153	0.3144±0.3939	0.0317±0.0298	2059.98±1409.36	36.78±30.59	11.51±7.76	30.94±8.40
Female	0.3386±0.1217	0.5482±0.2780	0.0208±0.0139	1947.71±875.76	33.07±20.45	8.24±4.11	33.81±9.98
Gestational age							
< 30 wks	0.3083±0.1215	0.4731±0.5614	0.0191±0.0181	2093.64±634.11	43.81±31.58	13.35±11.00	29.83±8.08
≥ 30 wks	0.3142±0.1172	0.3485±0.2711	0.0325±0.0285	2006.91±1448.18	32.87±27.04	9.53±5.22	32.59±9.12
Birth weight							
< 1200 g	0.2840±0.1026	0.3816±0.4850	0.0243±0.0257	2540.24±1464.13	46.12±31.70	11.47±9.05	31.51±10.31
≥ 1200 g	0.3378±0.1247	0.3920±0.2823	0.0324±0.0271	1583.62±919.80	26.75±21.47	9.72±5.04	32.02±7.57

From these data could be reported as followings. Firstly, the values of V_d/F, k_a, and C_{max} were higher in the female subject than the male; whereas, the k_e, AUC, $t_{1/2}$, and t_{max} values were higher in male subject than the female subject. Next, the inverted relationship between four pharmacokinetic parameters (k_e, AUC, $t_{1/2}$, and t_{max}) and GA was found. These values were higher in GA less than 30 wks. However, the directed relationship between the last three pharmacokinetic parameters (V_d/F, k_e, and C_{max}) and GA were observed. The more GA of the subjects, the higher values of these parameters. Lastly, the pharmacokinetics parameters categorized by birthweight showed the same trend as gestational age except k_a value which was higher in birthweight more than or equal to 1200 g. Although there were differences of these pharmacokinetic parameters for each factors, all of these differences were not reached statistically significant.

Validation of HPLC method

1. Linearity

The standard curve for plasma assay was conducted by spiking five ibuprofen concentrations into drug-free plasma and the average (n=2) peak area ratio of ibuprofen to mefenamic acid was calculated as presented in table 59and figure 11.

Table 59 Peak area ratio of ibuprofen to mefenamic acid at six different concentrations in drug-free plasma

Ibuprofen concentration (mcg/ml)	Peak area ratio (first time)	Peak area ratio (second time)	Average peak area ratio
0	0	0	0
4	0.0138	0.0127	0.0133
12	0.0473	0.0469	0.0471
25	0.1015	0.1023	0.1019
50	0.2153	0.2124	0.2139
100	0.4145	0.4131	0.4138

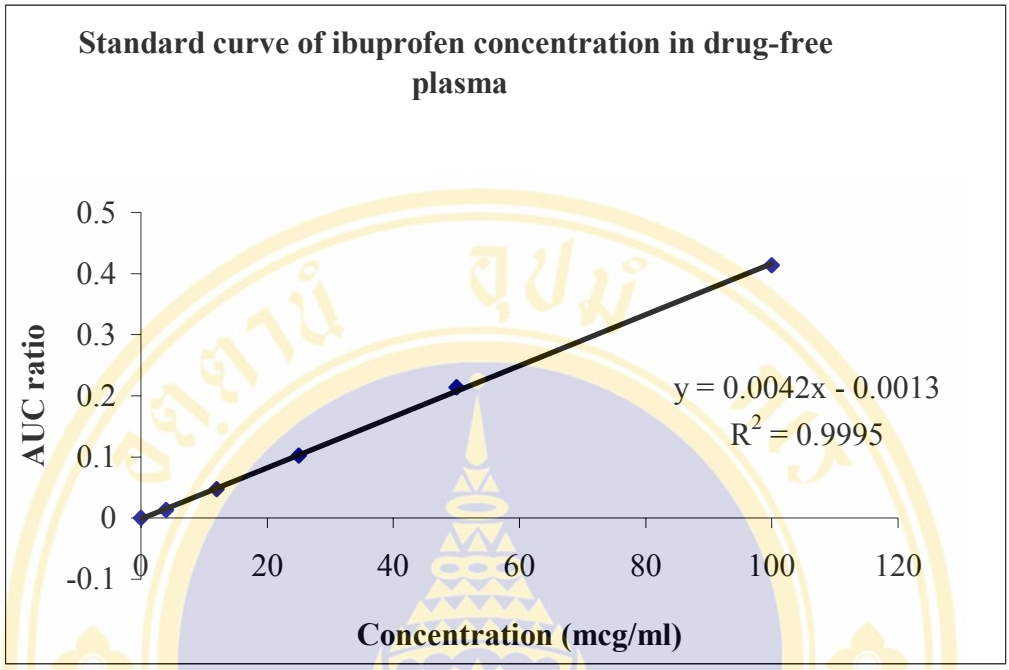

Figure 11 The standard curve of ibuprofen in drug-free plasma

The standard curve showed a good linear relationship with a correlation coefficient (r^2) of 0.9995 and the regression equation was: y = 0.0042x – 0.0013 where X is ibuprofen concentration in μg / ml and Y is the peak area ratio of ibuprofen to mefenamic acid.

2. Precision and accuracy

The intra-day assays of reproducibility and accuracy of the analytical method were performed. The results could be summarized as following table.

Table 60 Intra-day reproducibility and accuracy of the HPLC assay for plasma ibuprofen

Added conc	Measured concentration (mcg/ml)							%CV*	%Bias**
(mcg/ml)	First	Second	Third	Fourth	Fifth	Sixth	Mean±SD		
5	5.7914	5.262	5.7237	5.4581	5.7266	5.7266	5.6147±0.2084	3.7117	12.294
20	20.3922	20.1096	20.188	20.2988	20.1054	20.1719	20.2110±0.1132	0.5601	1.055
40	39.583	39.6983	39.8272	39.8165	39.9561	39.7986	39.7986±0.1378	0.3462	-0.5035
75	74.6703	74.8496	75.8574	75.2486	74.8524	75.5283	75.1678±0.4605	0.6126	0.2237

* %CV (percent coefficient of variation) = (SD * 100) / X

**% Bias = $\frac{\text{(Measured concentration – Added concentration)}}{\text{Added concentration}} * 100$

We measured four different concentrations of plasma ibuprofen. The calculated percent coefficient of variation (%CV) ranging from 0.3462 to 3.7117 and the calculated percent bias ranging from –0.5035 to 12.2940. All of these values were within acceptable ranges.

The inter-day assays were determined by measuring the same four concentations as intra-day asay on five different days. The results could be shown as following.

Table 61 Inter-day reproducibility and accuracy of the HPLC assay for plasma ibuprofen

Added	Measured concentration (mcg/ml)							%CV	%Bias
(mcg/ml)	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Average		
5	5.1851±0.1382	5.5942±0.1389	5.3728±0.2071	5.7992±0.0556	5.4995±0.1608	5.5853±0.0830	5.5702±0.1708	3.0661	11.4042
20	20.2827±0.3041	20.3450±0.4369	20.8394±0.8824	21.5331±0.3774	20.7391±0.3927	19.9322±0.2510	20.6778±0.7024	3.3967	3.3888
40	40.3252±0.2590	40.4886±0.7124	40.0163±0.2984	39.7766±0.7700	39.8116±0.8860	39.6945±0.4492	39.9575±0.6023	1.5073	-0.1063
75	75.7243±0.3981	75.4187±0.3667	75.5969±0.2902	75.4300±0.3153	75.1809±0.8580	75.5793±0.2797	75.4462±0.4407	0.5841	0.5883

The inter-day precision was demonstrated by %CV ranging from 0.5841 to 3.3967 and the accuracy by %bias ranging from –0.1063 to 11.4042. All of these values were within acceptable range.

3. Stability

Stability of three different concentrations of plasma ibuprofen was tested by freezing these samples in –60° scientific refrigerator at four different weeks. Freshy prepared blood samples (week 0) were assumed to be 100% added concentrations. The percent ibuprofen remaining at four different weeks were illustrated as following table.

Table 62 Stability of ibuprofen in FFP at four different weeks.

Prepared conc (mcg/ml)	Measured concentration* after each week			
	Week 1	Week 2	Week 3	Week 4
15	15.0569±0.0785	15.3520±0.0666	15.4596±0.3334	15.4812±0.3278
30	30.3269±0.2683	30.1866±0.4601	29.8615±0.6358	29.7488±0.4418
60	59.8766±0.6022	59.9326±0.6601	59.7337±0.5817	59.6016±0.3422

* Measured in six times.

Table 63 Percent of ibuprofen remaining in FFP at four different weeks

Prepared conc (mcg/ml)	% Ibuprofen remaining after each week				
	Week 0	Week 1	Week 2	Week 3	Week 4
15	100.00	100.38	102.35	103.06	103.21
30	100.00	101.09	100.62	99.54	99.16
60	100.00	99.79	99.89	99.57	99.34

The measured concentrations in each weeks were nearly equal to the initial concentrations so we could state that ibuprofen was stable through one month if the plasma was frozen at -60 °C before measurement.

CHAPTER 5

DISCUSSION

Clinical Efficacy

The primary objectives of this study were to determine whether three doses orally administered ibuprofen is efficacious in PDA prophylaxis in premature neonates with birthweight less than 1500 g and gestational age less than 34 weeks with minimal renal and hematological side effects and to study the basic pharmacokinetic parameters that might be related with PDA closure. Our results showed the higher percentage of symptomatic PDA reduction at day 3 in the study than in the control group (95.45% vs 65.00%; p = 0.018). Additionally, ibuprofen could reduce the incidence of silent PDA as the lower incidence of silent PDA found in the study group than in the control group (93.3% vs 64.3%; p = 0.014). The high efficacy of PDA reduction in this study went along with previous studies (28, 39, 40, 42).

The efficacy of ibuprofen for PDA prophylaxis has long been studied mostly in form of intravenous (28, 39-40, 42). These studies showed that the rate of ductal closure in the ibuprofen group was higher than in the control group with minimal renal side effects. However, intravenous ibuprofen was not as widely available as the suspension form in many developing countries including Thailand. If the suspension form has the same efficacy as the intravenous form, it will be in practical use because it is now widely used for other indications. Hariprasad et al.(41) administered oral ibuprofen (10 mg/kg at first dose and 5 mg/kg at 12 hourly interval) to 13 premature infants whose PDA were closed in 11 (84.62%) with no oliguria or bleeding tendencies. The high proportion of PDA closure without renal and hematological side effects was like our study but his report was only his own experience. His study differed from our study in term of the lower dose and shorter dosage interval of the second and the third dose, smaller sample size, and uncontrolled trial. Although there were different aspects from our study, his study reported the possibility of using oral

ibuprofen as an alternative for PDA treatment. Recent study by Heyman et al.(86) in 22 premature neonates reported the equal efficacy (95.45%) and low renal side effect of ibuprofen suspension as our study. His study was very similar to ours; for example, subject number, numbers of dose and dosage interval. However, no echocardiography was performed after each dose in our study and the GA and BW of our study were a little bit higher. From our and Heyman (86) studies, we may state that ibuprofen suspension have high efficacy in both prophylaxis and treatment of PDA as shown by equal proportion of closed PDA subjects (21/22). Our results also suggested that ibuprofen could reduce the incidence of silent PDA but more studies are needed to confirm this result.

In Thailand, two studies by Phatcharaphun et al (43) and Suravetwongphat et al (47) that had study designs closed to our study were conducted. The results showed high incidence of PDA reduction similar to our study. The unchanged renal function reported by Saowanee (43) was similar to our study but in Suravetwongphat's study (47), about 25% showed transient oliguria and serum creatinine rising but returned to normal later without intervention. Although in Suravetwongphat's study there were some rising in serum creatinine, the increased level was much lower than those with indomethacin treatment. The study reported about 5% gastrointestinal bleeding which was nearly equal to the incidence in our study. There was higher percentage of gastrointestinal bleeding in the study than in the control group despite normal platelet count and INR. In preterm neonate, the mucosal barrier was not fully developed (77, 98) so they could be prone to gastric irritation by many non-steroidal anti-inflammatory drugs. The high percentage of bleeding disorder could affect the other GI side effects such as higher incidence of abdominal distension, feeding difficulties, NEC, time to start feeding and reach full feeding. All of these differences in gastrointestinal problems didn't reach the statistically significances except for the time to start feeding which corresponds with the previous study (40). Previous studies had been shown that ibuprofen administration with milk could reduce local gastrointestinal side effects without interference on absorption rate.

Although Cooper Peel (37) and Ahlfor (82) reported that ibuprofen may cause bilirubin encephalopathy, the level from the suspension form in this study might not be

high enough to cause this effect. We didn't evaluate the bilirubin effect in this study. More studies to confirm this result may be necessary.

The duration of hospitalization tended to be longer in the study than in the control group. This could be explained by the fact that physicians had to waste more time to treat clinical sepsis and gastrointestinal bleeding until clinical conditions were stable.

The incidence of BPD in the study group was higher than in the control group which could be explained by the longer duration of oxygen therapy, mechanical ventilator usage and higher percentage of sepsis. There were no cases of PPHN in this study as reported by Gournay (38). The enrolled cases were more mature and the time of drug administration was slower than that study. Consequently, the normal decrease in pulmonary vascular resistance might not be disturbed.

In this study, the re-opening of PDA in each study group after completion of drug administration could be observed. Almost all of the cases were associated with clinical sepsis. Infection had been suggested as one of the precipitating causes of PDA re-opening especially when *Candida sp.* and gram negative organisms were encountered (94). The assumption is that there were changes in 6-keto $PGF_{1\alpha}$ and TNF_{α} levels in infection state. The high level of 6-keto $PGF_{1\alpha}$ that was the urinary metabolite of PGI_2 could be metabolized to 6-keto $PGF_{1\alpha}$ which is a strong ductus arteriosus-relaxing agent like PGE_2 (94). TNF_{α} may also affect ductal relaxation either directly or indirectly (94). We didn't measure the level of these mediators.

Pharmacokinetics

From the plasma drug concentration data of 22 subjects in the study group, only seventeen subjects were able to fit the Win Nonlin® program. The pharmacokinetic parameters of oral ibuprofen among the preterm infants with PDA showed a large interindividual variability.

The Pharmacokinetics of oral ibuprofen suspension in twenty-two premature infants in study group is also discussed. We couldn't fit drug data of five subjects to Win Nonlin program because there were much variations in pattern of drug kinetics. The pattern of drug absorption of the first two subjects couldn't predict the time to

reach maximum concentration and terminal slope of drug elimination because the sampling point of the first hour was very frequent. In adult and children, the time to reach maximum concentration is not longer than about one hour. This could be confirmed that the patterns of drug absorption and elimination in premature neonates differed substantially from older children (56-58, 80, 85) and adult (99) in the longer absorption and elimination phases. The last three subjects couldn't predict the pattern of drug elimination phase obviously in the first 24 hours because the drug levels of these subjects were constantly before administration of the second dose. Furthermore, the drug levels of some subjects were very fluctuation during the first 24 hours. All of these three subjects couldn't predict the terminal slope from their data so they couldn't be fitted to the Win Nonlin program for calculating Ke that was one of the essential initial parameters. In brief, only seventeen subjects were selected by the program to calculate three initial parameters. These were Vd/F, Ka, and Ke. The three values were used to calculate the secondary parameters.

The validation of HPLC method in this study showed good results. Firstly, the ibuprofen peak could be separatged obviously from the internal standard peak at the retention time about 8 min and 10 min, respectively so no interference peak such as the plasma component could be observed. Second, the standard curve between spiked ibuprofen concentrations and internal standard peak area ratio showed the good linear relationship with correlation coefficient of 0.9999. Thus we are sured that the measured drug concentration could be calculated correctly from the standard curve. In this study we couldn't use plasma from individual subjects as the limitation in sample amount but we assumed that the component of adult FFP was not differed from preterm FFP.Then the repeating measurement values in each time were not different as reflected by the acceptable ranges of %CV and %bias of the intra-day assessment so the measurement method has the good precision and accuracy. Next, the measured concentration at different day didn't affect the precision and accuracy of measurement since the %CV and %bias were within acceptable ranges. In other word, the time factor in each day didn't affect the HPLC process in this study. Lastly, the drawn blood samples had a good stability when they were centrifuged following by freezing at -20^{o} to -60^{o} immediately. If they were analyzed within one month, the drug in plasma didn't decompose as validated by the nearly equal drug concentration in each

week. All of these good validation results could confirm that the large variability in drug concentration between each subjects were not caused by the HPLC method.

There were an obvious large interpatient variability in k_a , k_e , AUC, T1/2, and t_{max} and some extreme values could be observed. We excluded these outliers by statistical method and reported the median value instead of the mean value because these values showed right skewness distribution. The values of Cmax, Tmax, T1/2, and AUC of ibuprofen suspension were about two to three times higher than the previous study (81) but the half-life value was almost as equal as the value shown by the previous studies (26, 84). This could reflect that the elimination of ibuprofen in these premature infants was very slower than the older children and adults. The Cmax and AUC values were much lower than reported by the previous studies (54-55) because an oral preparation had absorption variability higher than a parenteral one. Additionally, babies of smaller GA and BW tended to have longer half-lives and higher AUC values but the Cmax value was nearly equal to the other group like the Sharma's study (81). However; this was only the subgroup analysis and it didn't reach the statistically significant. The more sample sizes will be needed to draw the definite conclusion. Although this was only the report, we may explain the difference as a result of the immaturity of drug biodisposition process in smaller neonates. All of these variabilities may be explained by the differences in physiologic factors of premature newborns from older children and adult as the followings.

Firstly, the process of drug biodisposition could be affected by many physiologic change from PDA condition so some pharmacokinetic parameters may be changed in the following examples. The reduction of blood flow to drug-elimination organ could be resulted in the reduction of drug elimination from the body as reflected by the prolonged half-life value. Moreover, the higher TBW proportion in both preterm and full term neonates together with acidosis from PDA were the cause of high Vd (54, 94). Overmeire (54) reported that the Vd value could be reduced significantly after the PDA was corrected. This lead to improve renal perfusion to eliminate exceed extracellular water. The acidosis from RDS is also a precipitating factor for high Vd value in some drugs such as gentamicin (94). In conclusion, the major physiologic change from PDA condition is exceeding extracellular water, acidosis, and blood flow reduction. The major affected pharmacokinetic parameters

are Vd, CL, and T1/2. One of th explanations for the variability in Vd and T1/2 values in this study might be resulted from this phenomenon.

Then the change in gastric emptying time could affect the drug absorption process. The gastric emptying time was delayed immediately 10-20 hours after birth in both full term and preterm infants (61-62, 76). It will reach to the adult value at the age about 6-8 months (61-62, 76). One study (76) reported that RDS in premature infants could prolong the gastric emptying time in premature infants (76). Additionally, the peristalsis pattern in preterm neonates was unpredictable and couldn't relate this factor with GA. In this study, the absorption pattern from most subjects were slowly so the Ka and Tmax values were rather high. In short, the cause of these variabilities may be beneficial for sustaining therapeutic level that necessary for PDA closure. However; other factors such as metabolism and elimination patterns should be considered.

Next the change in the activity of drug-metabolizing enzymes in premature neonates could affect the ibuprofen metabolism. The major pathways of ibuprofen metabolism are hydroxylation and carboxylation which involved with the phase I and phase II enzymes. In phase I enzyme, the CYP2C9 activity in preterm neonates was very low after birth then the activity increases about 50% in the first week and it reaches adult level after the first year of life (76). In phase II enzyme, the UGT2B7 activity that is essential for metabolizing many NSAIDs including ibuprofen is only about 10-20% at 15-27 weeks fetal life and it reaches the adult value at 2 months to 3 years (93). The immaturity of the both phases enzymes may explain the reduced ibuprofen metabolism in premature neonates in this study. Additionally, genetics polymorphism of CYP2C9 genotype could affect the CYP2C9 expression activities. The intrinsic ibuprofen clearance was much higher for the CYP2C9 (Arg144) variant than for the CYP2C9 (Cys144) variant. Although our study didn't test the genetic variant for individual subject, we might assume that the long half-life value may be in part from the slow metabolizer group. Furthermore, these subjects group might be assumed to be prone to the gastrointestinal bleeding side effect from this drug.

Lastly, the immaturity of renal function in premature infants could markly affect the elimination of many drug including ibuprofen. The glomerular filtration rate (GFR) in premature infants is about two times lower than termed infants (76-77). The

development of GFR in premature infants is slower than term infants. In addition, the tubular function in premature neonates is more immature than term neonates and its development is slower than the GFR function. Aperia et al (93) also supported that postnatal renal function development during the first two days in preterm infants was less than term infant. The most preterm infants in our study were administered drug within 24 hours after birth that was faster than other studies so the kidney might not function completely. All of these above reasons could explain the prolonged half-life and the reduced clearance values in this study.

Recently, there were two studies (37, 82) reported that the high ibuprofen level could be competitive binding of albumin from bilirubin because this drug was highly protein bound so the free bilirubin could be increased. However, the drug level in this study was about three times lower than the previous studies (54-55) that used intravenous form so the possibility to cause this phenomenon was low. Not any subjects in this study developed this side effect but this undesirable adverse effect should be awared and monitored in other subsequent studies.

Pharmacokinetic-Pharmacodynamic relationship

Although there were no studies directly reported the ibuprofen level that may be necessary for PDA closure, there was a study (86) that reported the pharmacokinetic parameters of this drug in premature neonates. The Cmax, Tmax, and T1/2 values from Sharma et al (86) were rather lower than our study. These differences may be from many causes; for example, the time of drug administration in our study was less vary than Sharma's study (8.5 – 44.5 hrs and 4-72 hrs). The difference in physiologic factor in each day may affect the pharmacokinetic of ibuprofen. Next, the different method of HPLC used could be affected. Unlike the previous study (86), our study used internal standard to reduce the measurement error while the previous study (86) didn't. However, our study reported the drug level that may be necessary for PDA closure was lower than the reported level by Aranda (55) (180.6±11.1 mcg/ml) and Varvarigou (28) (180.6±50.8 mcg/ml). From these comparisons, the exact level for PDA closure still didn't know but its level might be lower than the average level from our study (Cmax = 31.78 mcg/ml). Interestingly,

one study (86) showed that 14/22 (63.4%) subjects closed their PDA with one dose of ibuprofen as proven by echocardiogram. It is possible that the slower rate of oral ibuprofen absorption, the longer time to reach Cmax, and the prolonged exposure time could sustain ibuprofen level long enough to exert its pharmacological effect at the ductal site.

According to the table 57, all 17 subjects were closed PDA at day 3 with the average maximum concentration 31.73 mcg/ml and drug concentration at before the second dose 29.14 mcg/ml (9.85 – 45.86 mcg/ml). However; the minimum drug concentration at 30 minutes before the second dose in eleven silent PDA subjects was 12.48 mcg/ml. This value was higher than the minimum values of the total 17 subjects.

In our study, there was only one subjects that PDA was still opening after completion of drug administration at day 3. We may explain that the time to maximum concentration of this subject was only 2 hours together with the rapid elimination from the body. Consequently, the drug level couldn't be sustained long enough for exerting its pharmacological action. The lowest drug concentration at 30 minutes before the second dose 7.37 mcg/ml supported the above assumption. From the time vs concentration pattern of this subject, we try to calculate the ka and ke values in order to predict other secondary pharmacokinetic parameters. By calculation, ka 4.3014, Vd/F 0.1949, ke 0.0999, t1/2 6.99, CL/F 0.0193, and AUC 517.6050. The highest ka value may explain the fastest absorption. The high ke with low t1/2 values lead to rapid drug absorption and elimination from the body, respectively. We assume that the function of gastrointestinal and renal systems were rather mature that was the specific characteristic of this infant although gestational age and birth weight were premature. Because his/her mother didn't receive dexamethasone that could reduce RDS but receive magnesium sulfate injection that could suppress respiratory tract of the infant, the hypoxic condition from these causes may prolong the contraction of the ductus arteriosus in this infant. Additionally, his/her mother presented urinary tract infection before laboring so infection could potentiate the presence of RDS. Lastly, we speculated that this infant has genetic polymorphism of CYP2C9 (Arg144) variant that could metabolize ibuprofen rapidly.

CHAPTER 6

CONCLUSION

In this study, with completion of three doses administration, ibuprofen suspension could reduce the incidence of PDA evaluated at day 3. Ibuprofen suspension didn't significantly affect the renal function and hematological laboratory value. Although hematological laboratory values were within the normal limit, the incidence of gastrointestinal bleeding was higher in the study than in the control group. Local gastrointestinal side effects warrant physicians if the drug were administered. To reduce these side effects, ibuprofen suspension might be administered with milk. The impact of BW and GA on closure of PDA was confirmed in our study. Subjects with lower birthweight or gestational age tended to have higher incidence of PDA.

There were high interindividual variabilities of pharmacokinetic parameters in premature infants. The Cmax value was nearly the same as that in older children or adult value. The t_{max} and $t_{1/2}$ were longer than those studied in older children or adult value. The differences could be explained by the immaturity of drug biodisposition organs in premature infants so that the processes of absorption, distribution, metabolism, and elimination could be affected. After completion of drug administration on day 3, twenty-one patients had closed PDA with Cmax about 30 mcg/ml. The exact level that could close PDA was unknown but from this data we can state that the level produced by suspension form with long elimination time may be adequate for PDA closure. The prolonged t_{max} and $t_{1/2}$ of the drug might be able to exert its fully pharmacologic action at the ductal site so that only single dose of ibuprofen may be adequate for closing PDA. This requires more studies. Data of drug concentration at each time points could be used to predict the drug concentration after administering the second and the third doses. The difference between average observed and predicted concentrations was not statistically significance. However; many changes in physiologic condition of premature infant during each day should be considered in the use of this predicted values. The values of AUC, $t_{1/2}$, and t_{max} tended

to be higher and C_{max} tended to be lower in male subject, GA<30 wks, and BW<1200 g. The differences were statistically significant and more studies are needed to evaluate whether gender, GA, and BW may affect the pharmacokinetic parameters.

Recommendation for further studies

From the result of PDA reduction and pharmacokinetics of oral ibuprofen suspension in premature neonates, we have some recommendations for other studies as the followings. Firstly, ibuprofen appeared to be advantage in silent PDA reduction on day 3 with statistically significant. If this is true, ibuprofen administration may be useful in prevention of both symptomatic and silent PDA in premature infants. Further studies might use this result to calculate enough sample size and design well randomized controlled trials to verify this advantage. If all of these benefits were proven by well designed randomized controlled trials, ibuprofen might be served as another standard treatment and prevention of PDA. Secondly, the prolonged elimination phase of ibuprofen may sustain the adequate drug level to close PDA. The subsequent studies may evaluate the efficacy of single dose ibuprofen administration in PDA closure in order to prove the benefit of prolonged elimination to achieve the maximum pharmacologic action at the ductal site and may evaluate the efficacy of ibuprofen in back-up treatment if the patients need more than one dose to treat PDA. Additionally, the single dose pharmacokinetics of ibuprofen should be studied concurrently. We suggest that the sampling time should be extended after twenty-four hours of drug administration in order to predict the terminal slope obviously and the calculated elimination half life will be more accurate. Lastly, the ongoing studies should be performed to correlate drug concentration level with PDA closure.

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44. อัญชลี ลิ้มรังสิกุล. การศึกษาเปรียบเทียบประสิทธิภาพและผลข้างเคียงของยา ibuprofen ชนิดรับประทานและยา indomethacin ในการรักษา PDA ในทารกเกิดก่อนกำหนด. วิทยานิพนธ์ตามหลักสูตรเพื่อวุฒิบัตรแสดงความรู้ความชำนาญในการประกอบวิชาชีพเวชกรรม สาขากุมารเวชศาสตร์ของแพทยสภา, 2544:1-26.

45. เกศนาถ จิรัปปภา. การศึกษาผลการรักษาโรคหัวใจ patent ductus arteriosus ในเด็กทารกคลอดก่อนกำหนดด้วยยา ibuprofen ชนิดรับประทานเปรียบเทียบกับยา indomethacin ชนิดฉีดทางหลอดเลือดดำ. วิทยานิพนธ์ตามหลักสูตรเพื่อวุฒิบัตรแสดงความรู้ความชำนาญในการประกอบวิชาชีพเวชกรรม สาขากุมารเวชศาสตร์ของแพทยสภา, 2545:1-38.

46. การเปรียบเทียบประสิทธิภาพของยาไอบูโพรเฟนและยาอินโดเมธาซินชนิดรับประทานในการรักษาโรคหัวใจดักตัสอาเธอริโอซัส โดย ร้อยเอก ประสาร เหมือนพงษ์ โรงพยาบาลพระมงกุฎเกล้า (บทคัดย่อ). วารสารกุมารเวชศาสตร์ ปีที่ 40 ฉบับที่ 1 มกราคม-มีนาคม 2544.

47. การศึกษาประสิทธิภาพและผลข้างเคียงของยา ibuprofen ชนิดรับประทานในการปิด PDA ในทารกเกิดก่อนกำหนด โดย นางสาวปิยนุช สุรเวทวงศ์ภาส มหาวิทยาลัยเชียงใหม่ (บทคัดย่อ). วารสารกุมารเวชศาสตร์ ปีที่ 41 ฉบับที่ 1 มกราคม-เมษายน 2545.

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APPENDIX

Observed ibuprofen concentration for individual subjects in the study group.

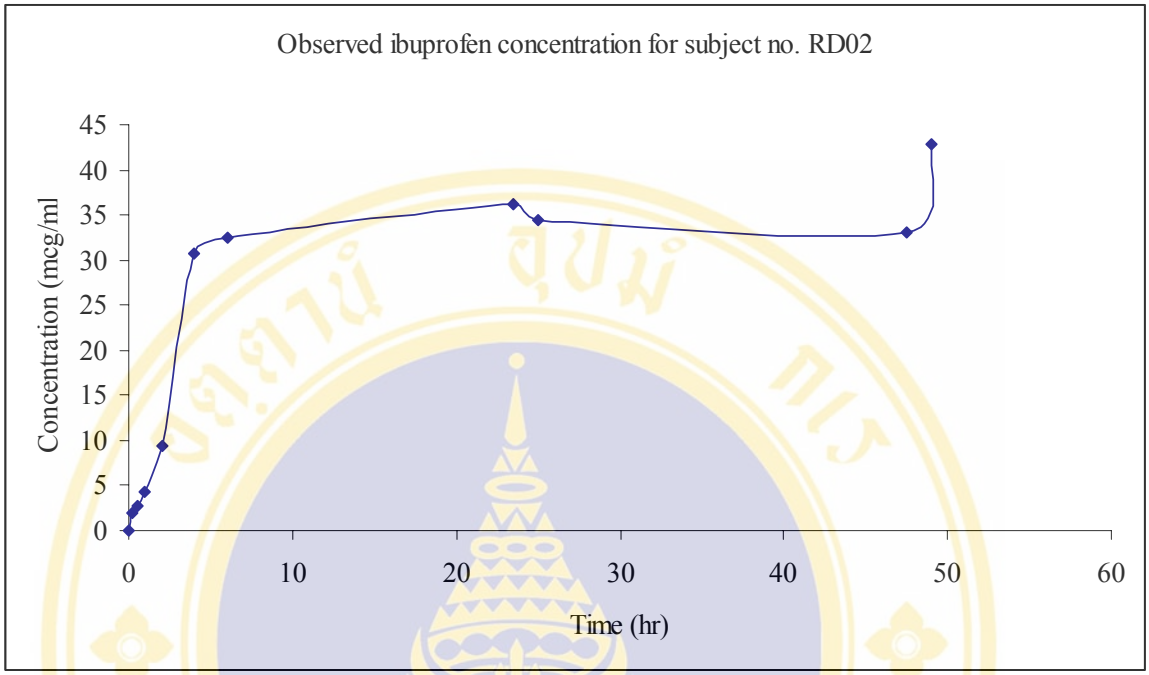

Figure 12 Plasma ibuprofen concentration in each sampling time points of the subject number RD02.

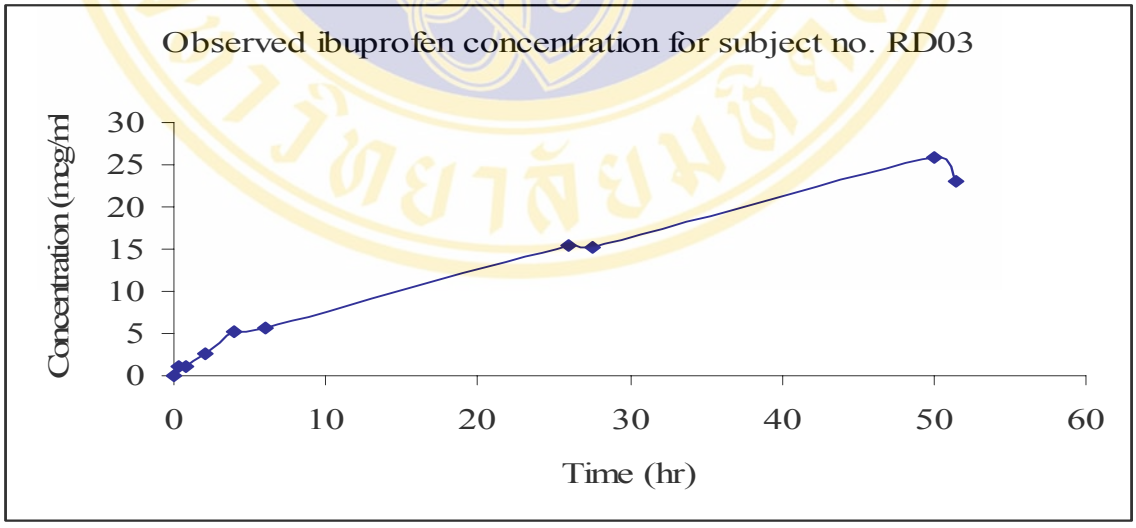

Figure 13 Plasma ibuprofen concentration in each sampling time points of the subject number RD03.

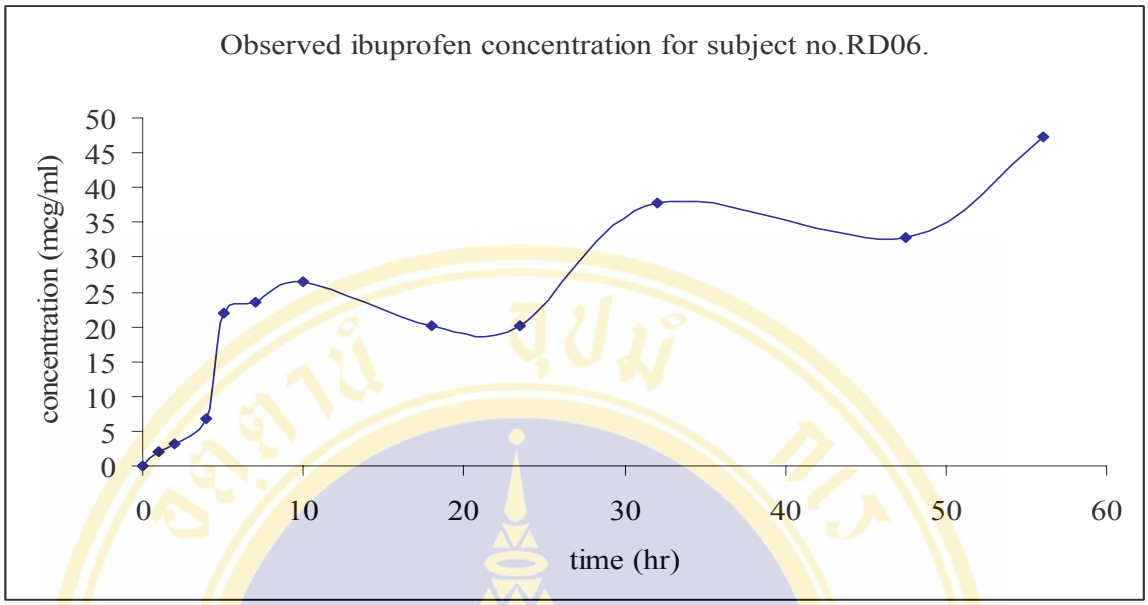

Figure 14 Plasma ibuprofen concentration in each sampling time points of the subject number RD06.

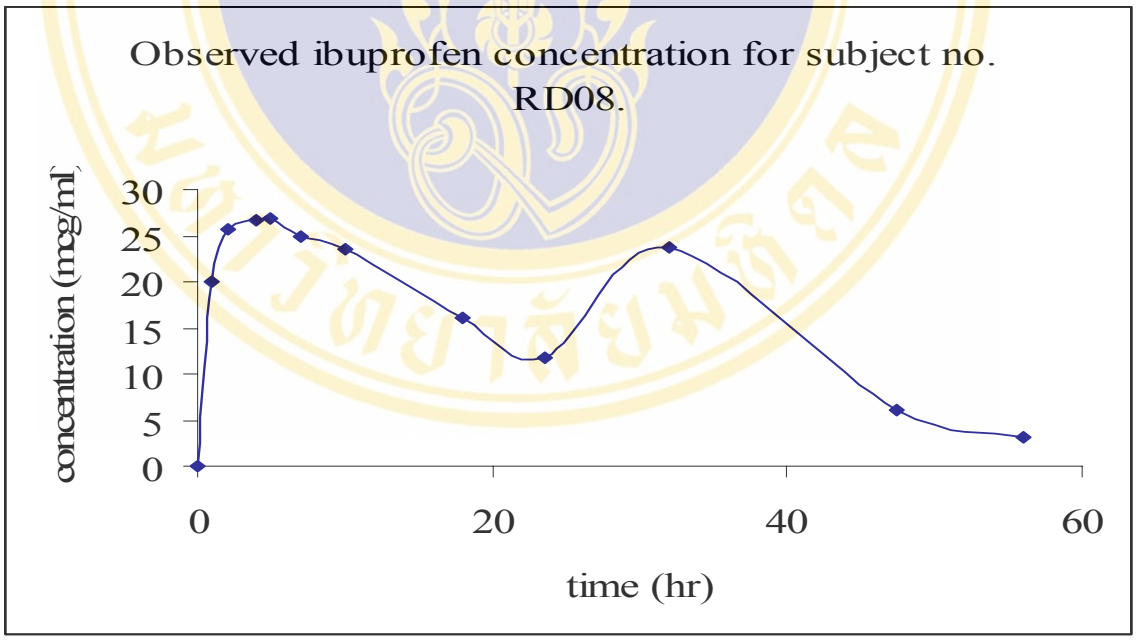

Figure 15 Plasma ibuprofen concentration in each sampling time points of the subject number RD08.

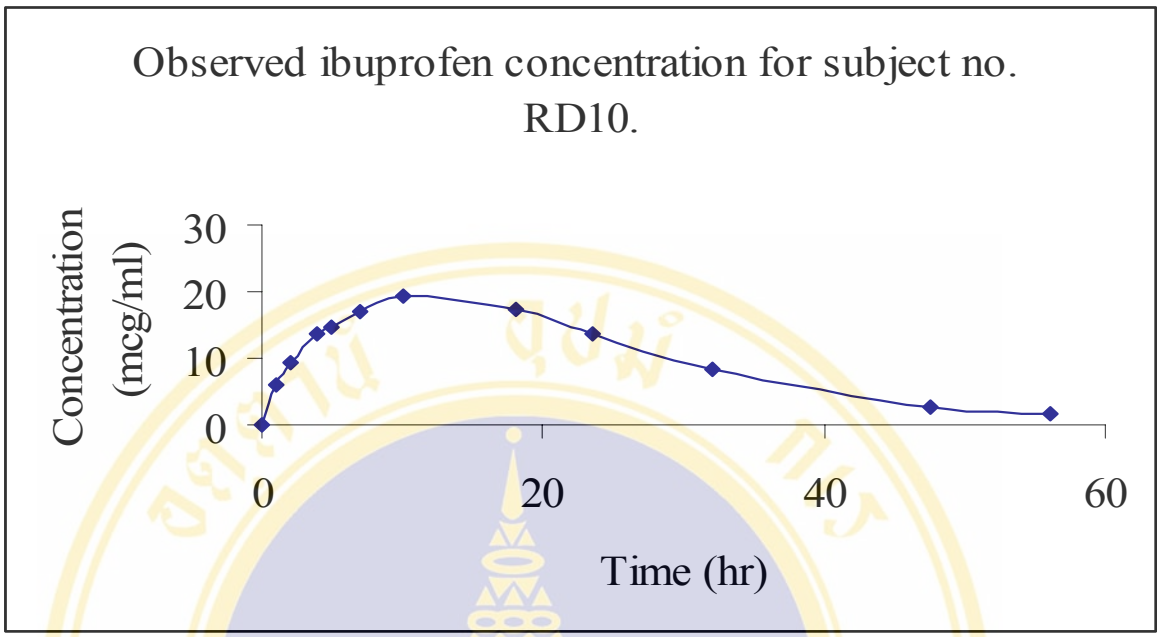

Figure 16 Plasma ibuprofen concentration in each sampling time points of the subject number RD10.

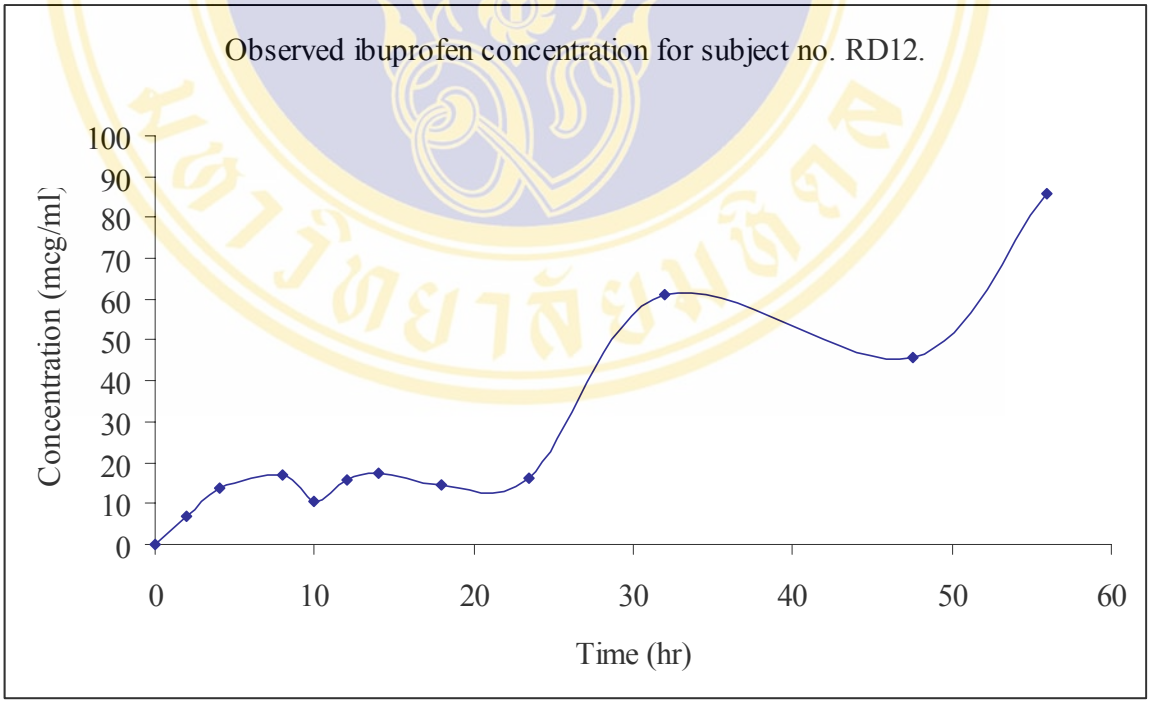

Figure 17 Plasma ibuprofen concentration in each sampling time points of the subject number RD12.

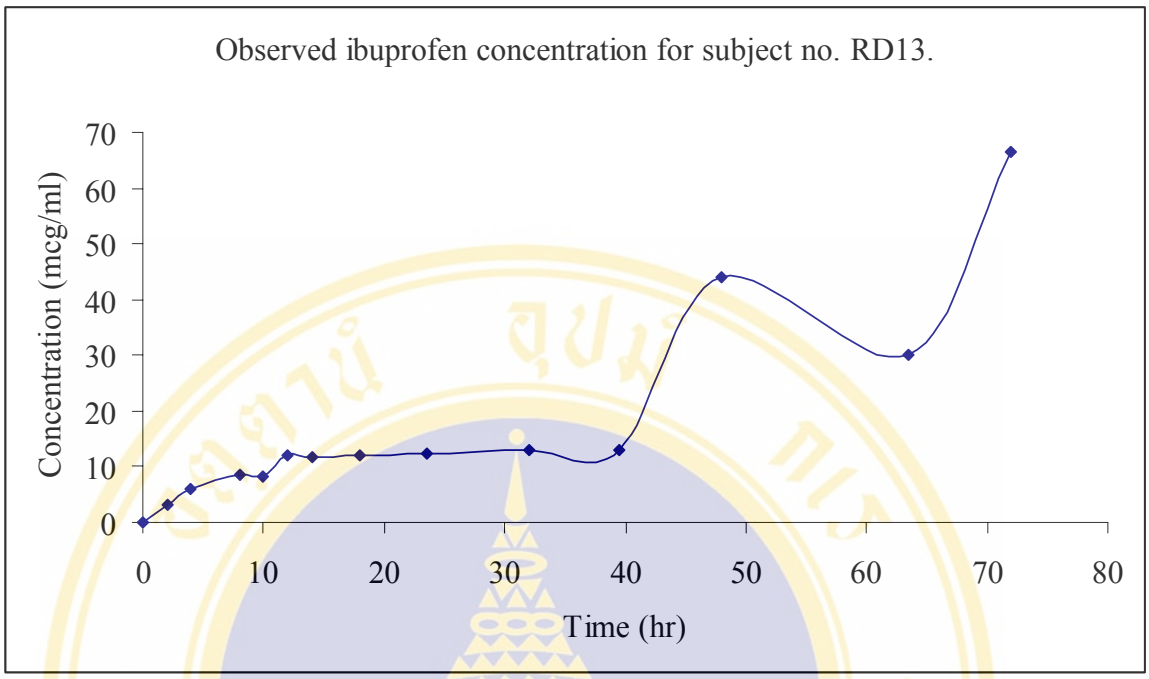

Figure 18 Plasma ibuprofen concentration in each sampling time points of the subject number RD13.

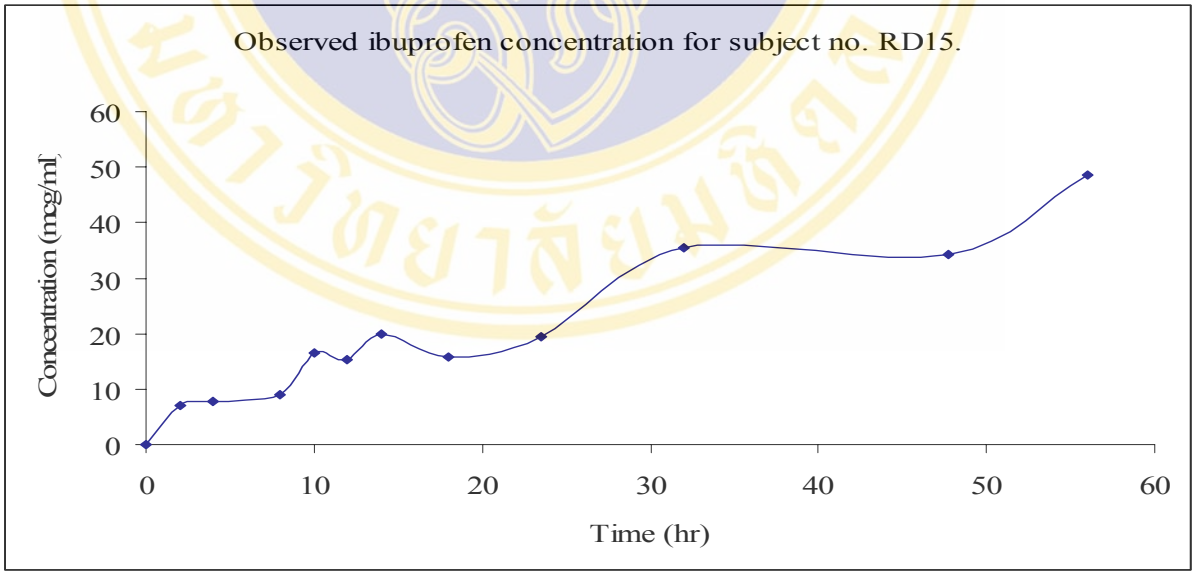

Figure 19 Plasma ibuprofen concentration in each sampling time points of the subject number RD15.

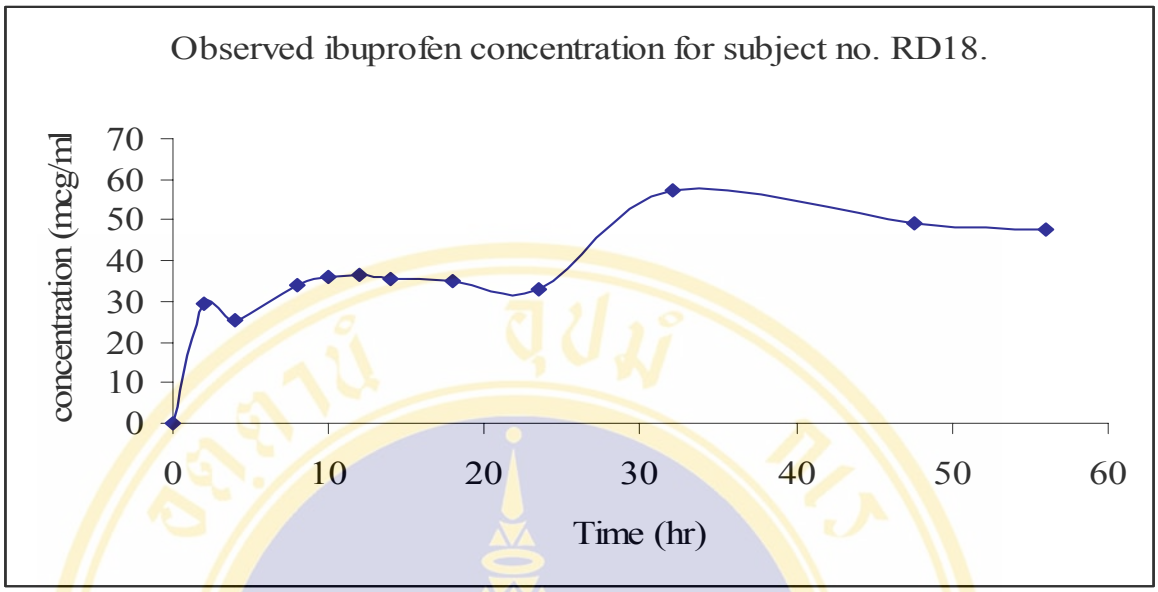

Figure 20 Plasma ibuprofen concentration in each sampling time points of the subject number RD18.

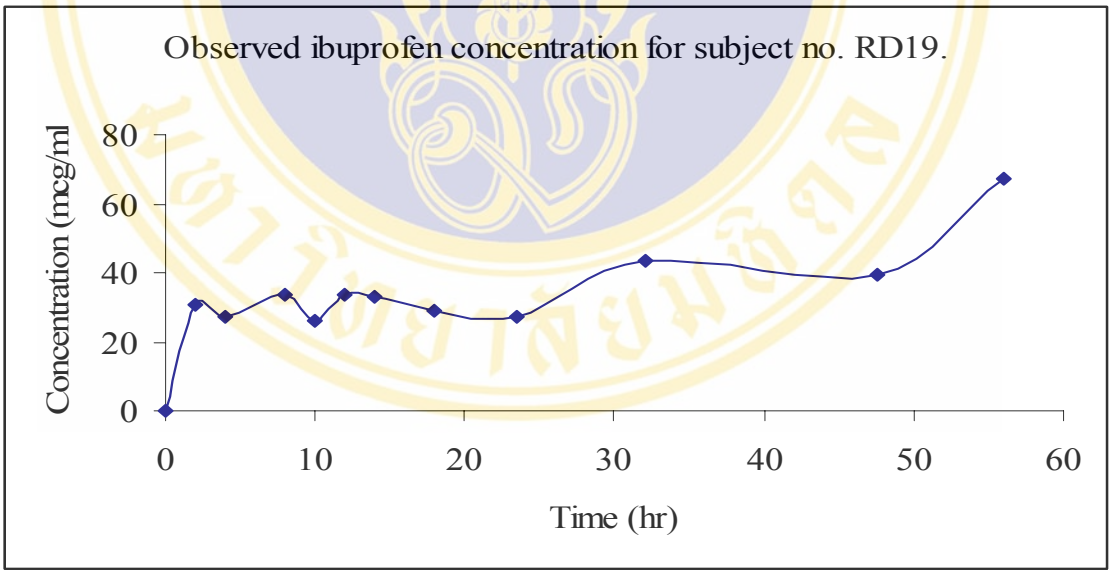

Figure 21 Plasma ibuprofen concentration in each sampling time points of the subject number RD19.

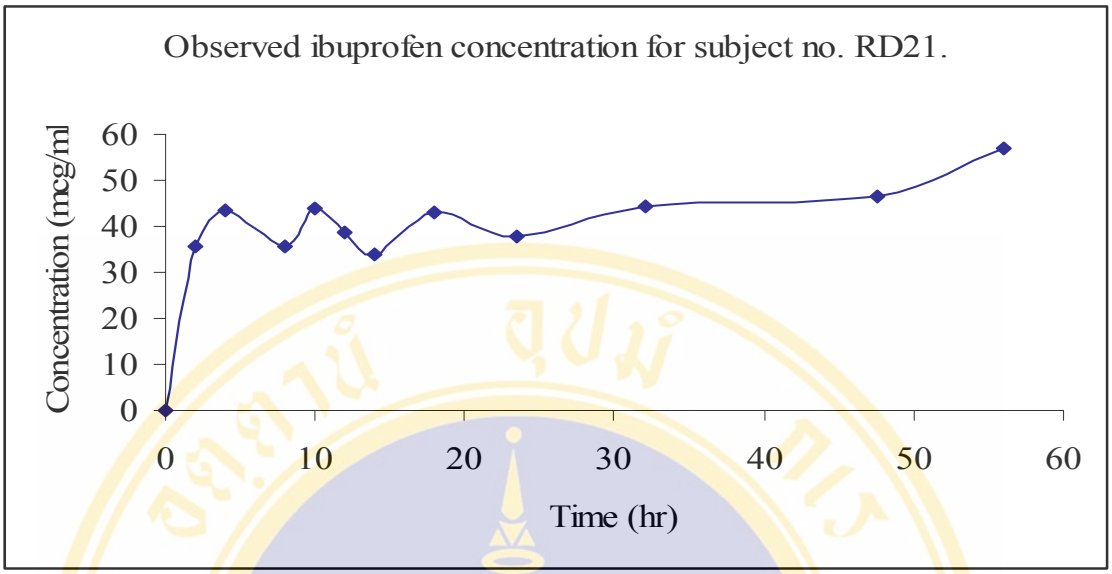

Figure 22 Plasma ibuprofen concentration in each sampling time points of the subject number RD21.

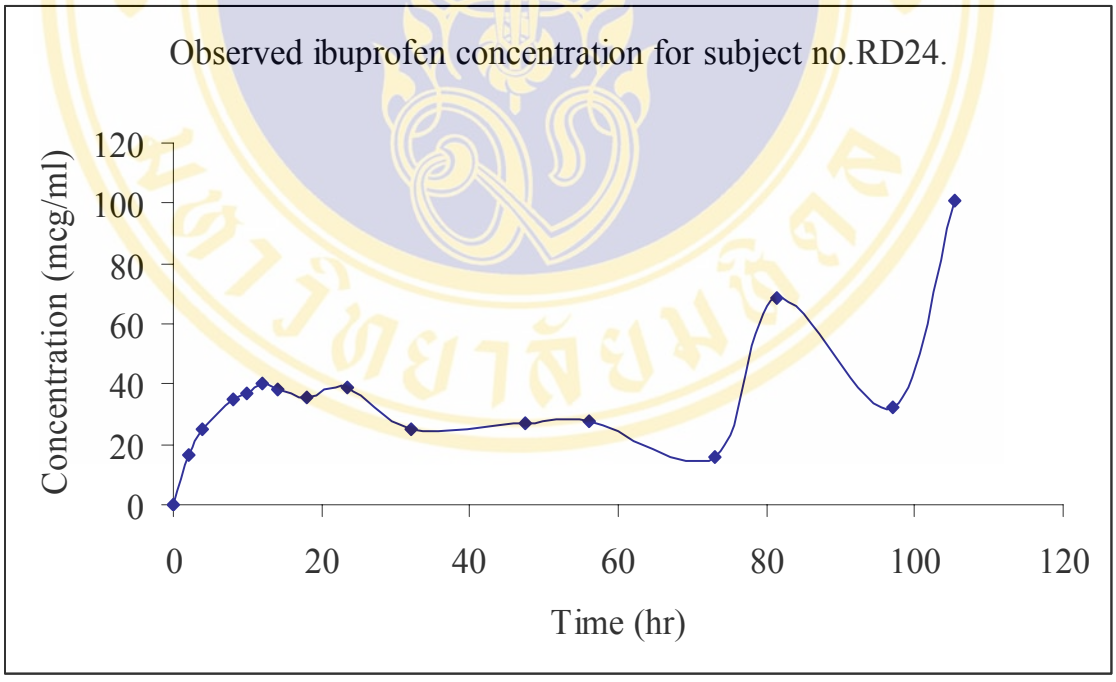

Figure 23 Plasma ibuprofen concentration in each sampling time points of the subject number RD24.

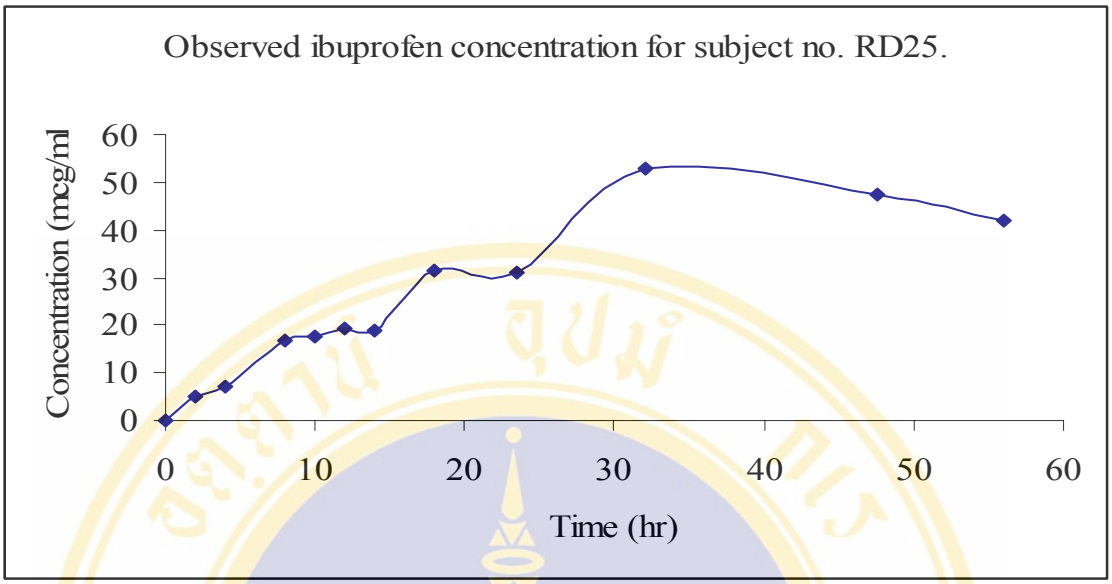

Figure 24 Plasma ibuprofen concentration in each sampling time points of the subject number RD25.

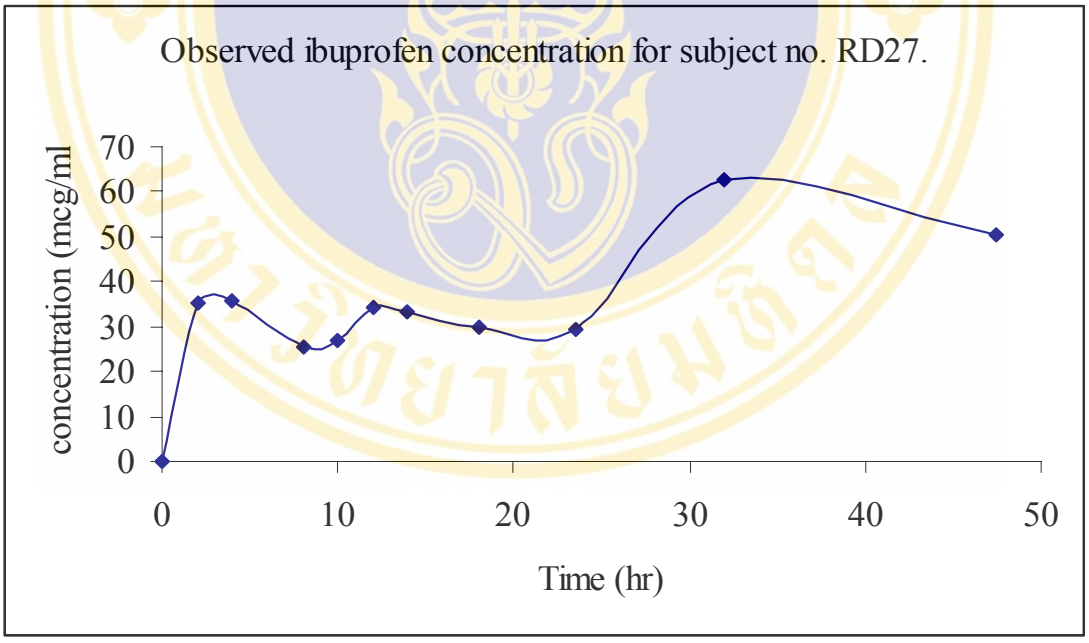

Figure 25 Plasma ibuprofen concentration in each sampling time points of the subject number RD27

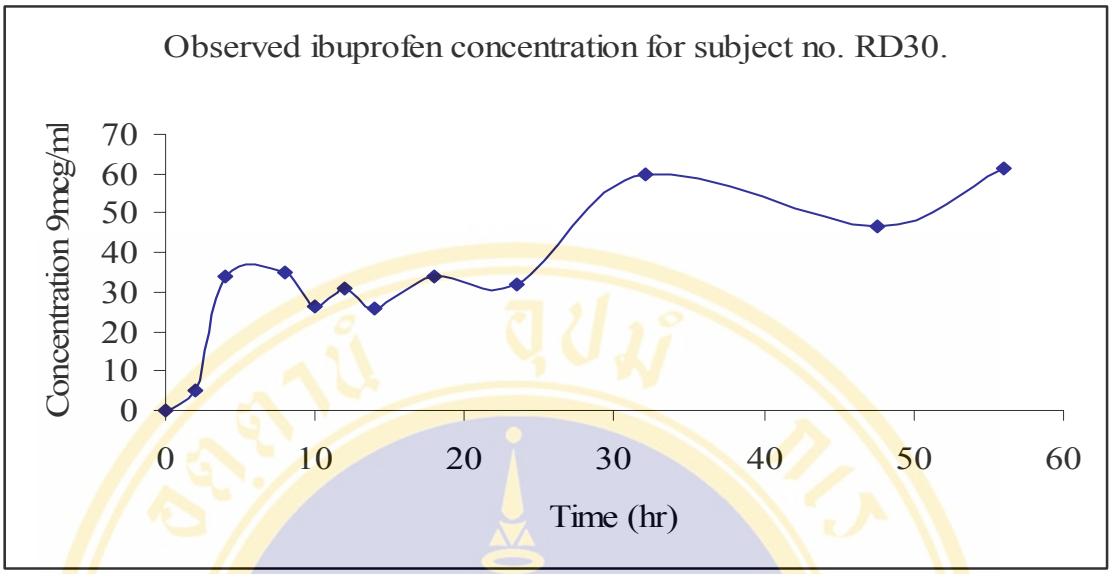

Figure 26 Plasma ibuprofen concentration in each sampling time points of the subject number RD30.

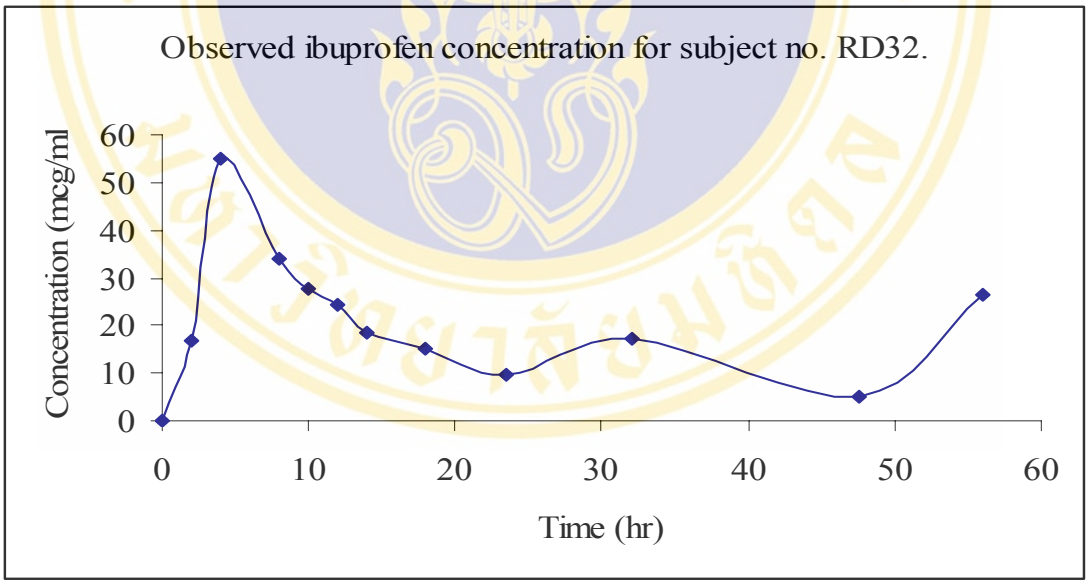

Figure 27 Plasma ibuprofen concentration in each sampling time points of the subject number RD32.

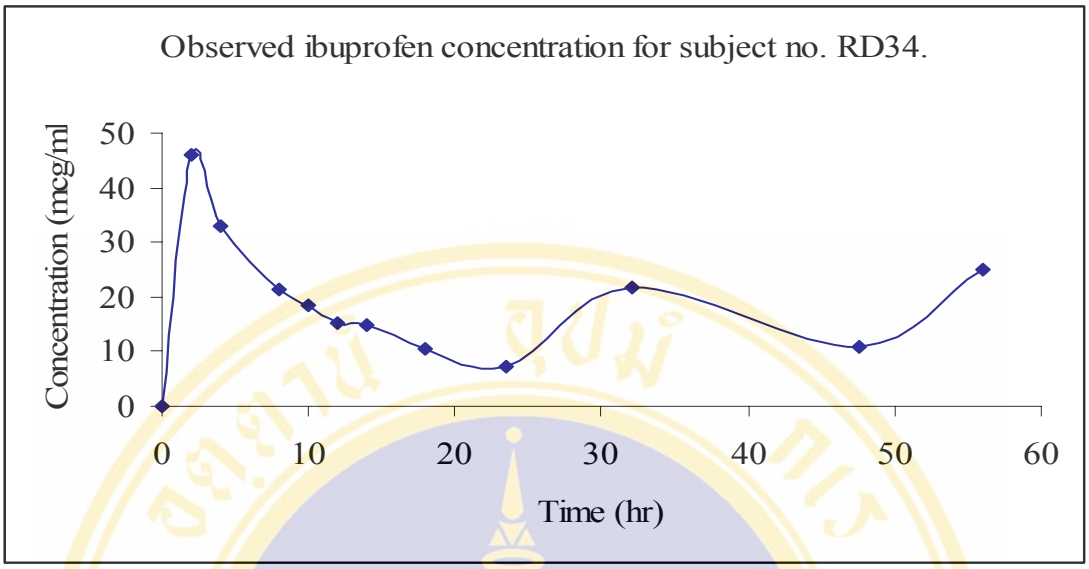

Figure 28 Plasma ibuprofen concentration in each sampling time points of the subject number RD34.

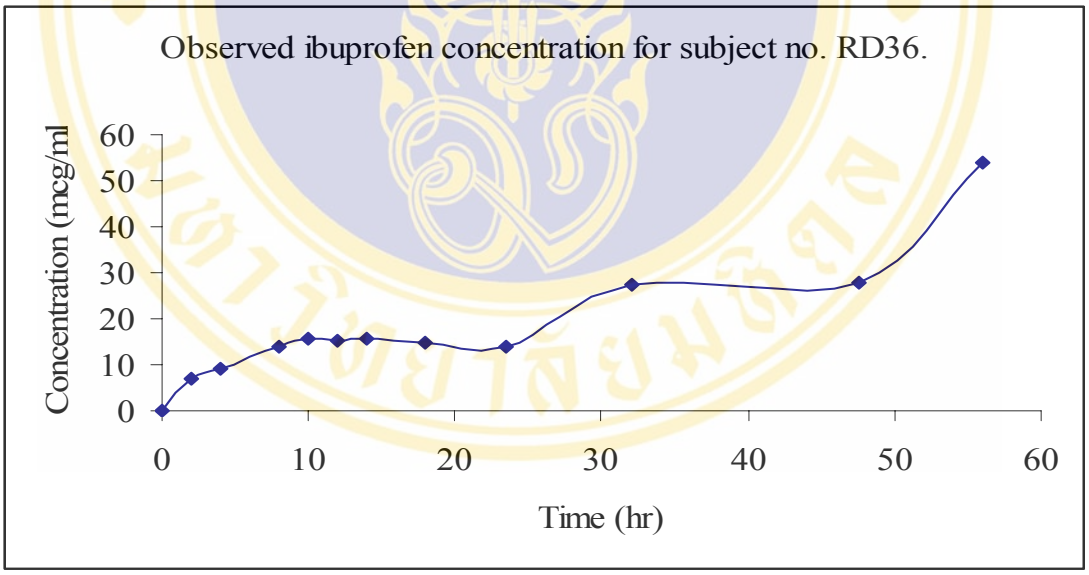

Figure 29 Plasma ibuprofen concentration in each sampling time points of the subject number RD36

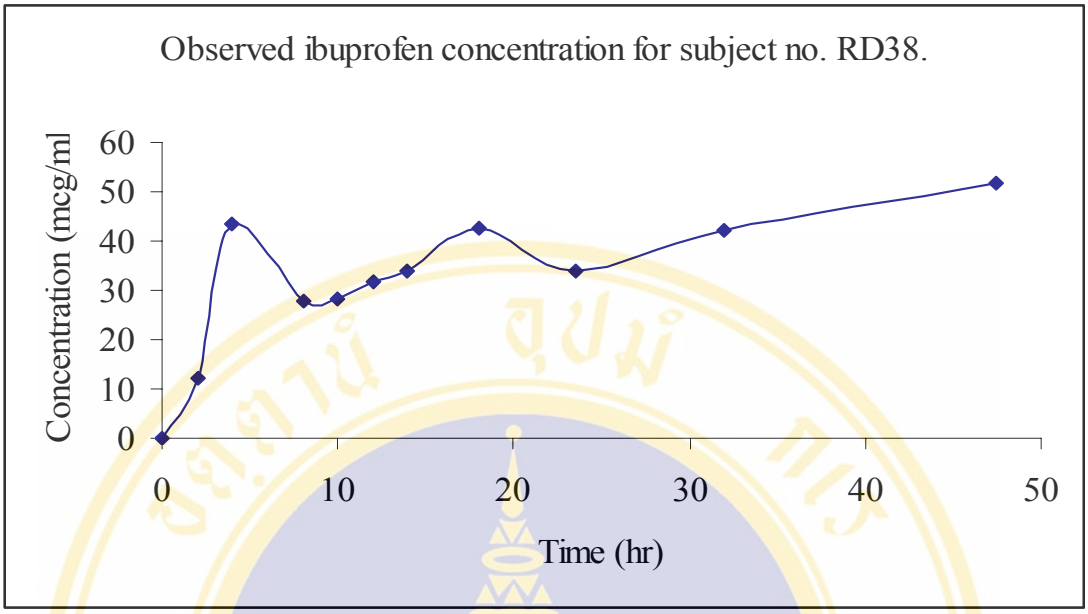

Figure 30 Plasma ibuprofen concentration in each sampling time points of the subject number RD38.

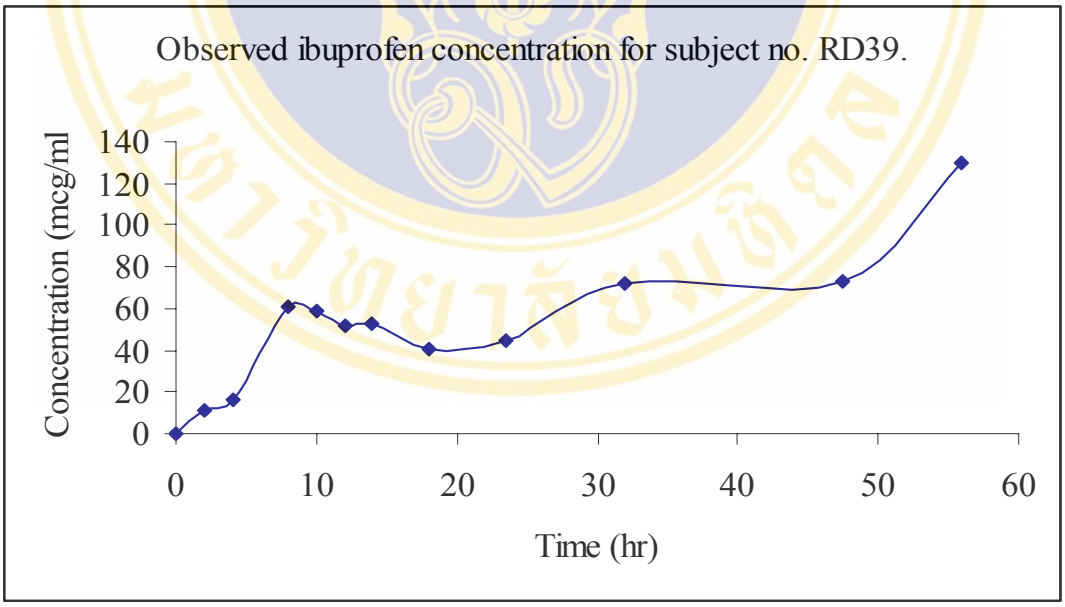

Figure 31 Plasma ibuprofen concentration in each sampling time points of the subject number RD39.

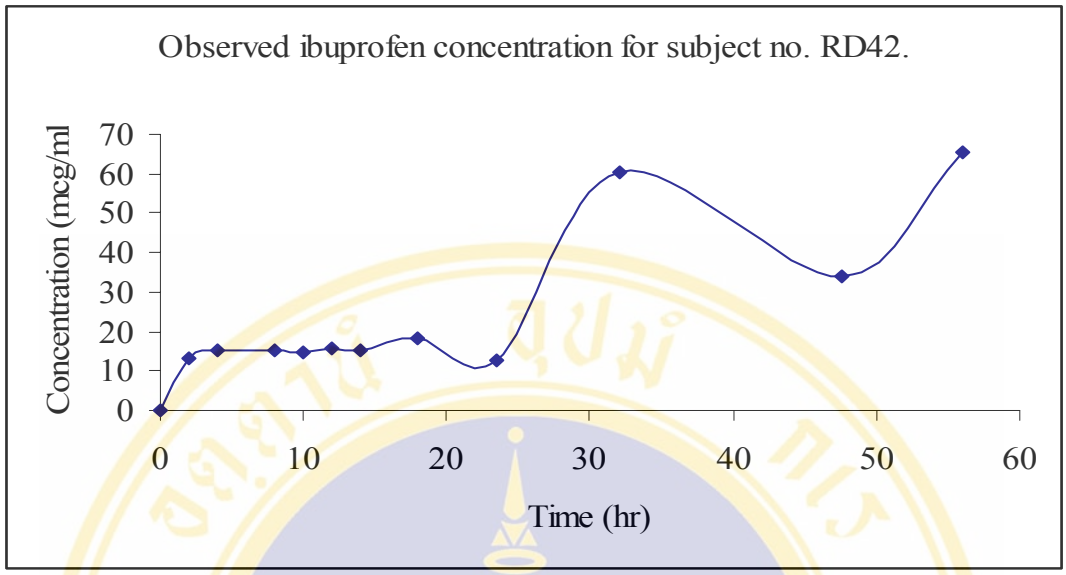

Figure 32 Plasma ibuprofen concentration in each sampling time points of the subject number RD42

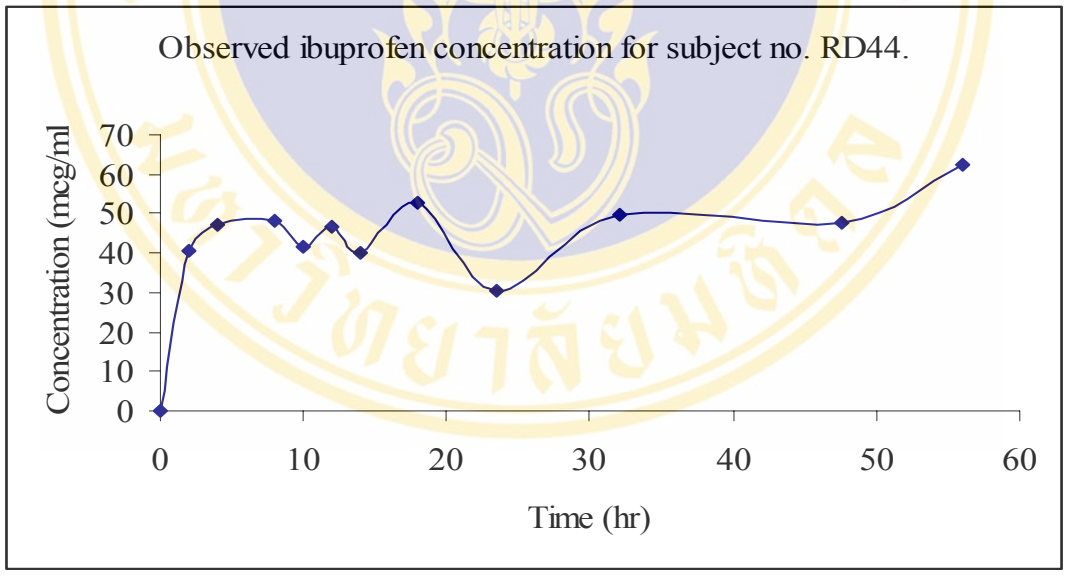

Figure 33 Plasma ibuprofen concentration in each sampling time points of the subject number RD44.

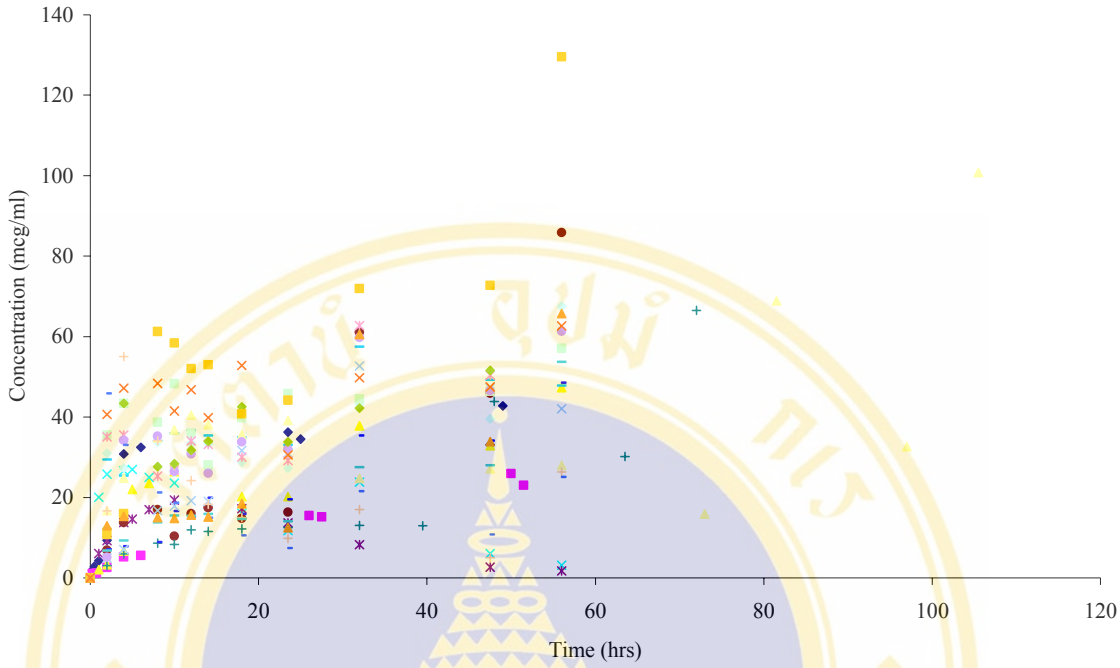

Figure 34 Spaghetti plot at every sampling time points for all 22 subjects in the study group.

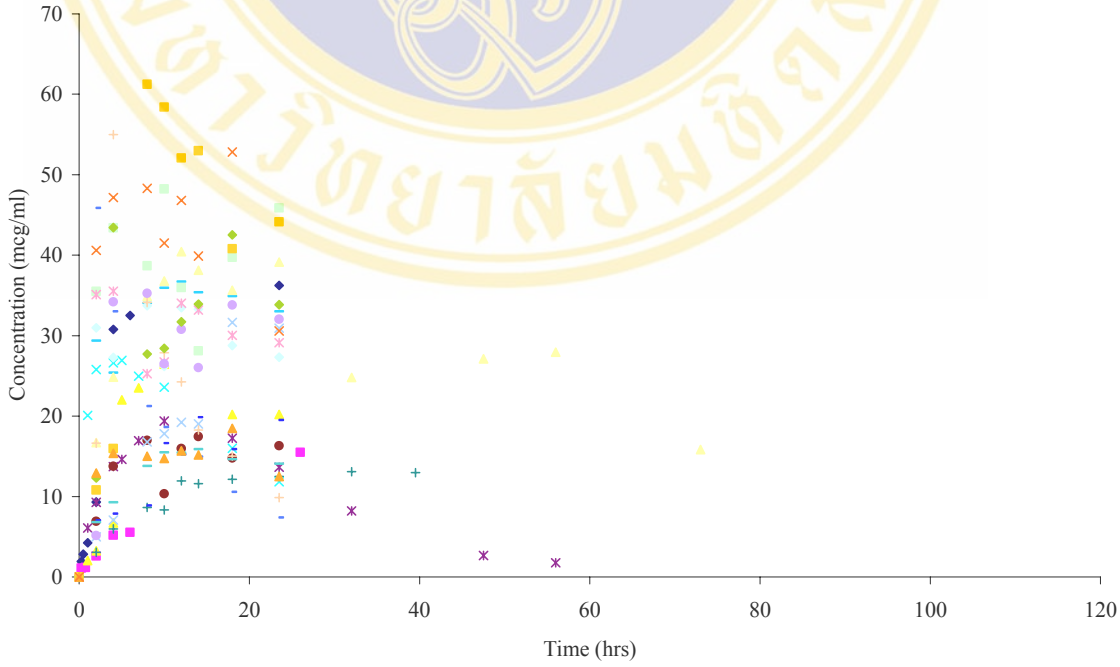

Figure 35 Spaghetti plot of single dose ibuprofen for all 22 subjects in the study group

Comparison of the observed and predicted ibuprofen concentration selected by Win Nonlin Program at each time point representing in both Concentration vs Time and Concentration (log scale) vs Time scales.

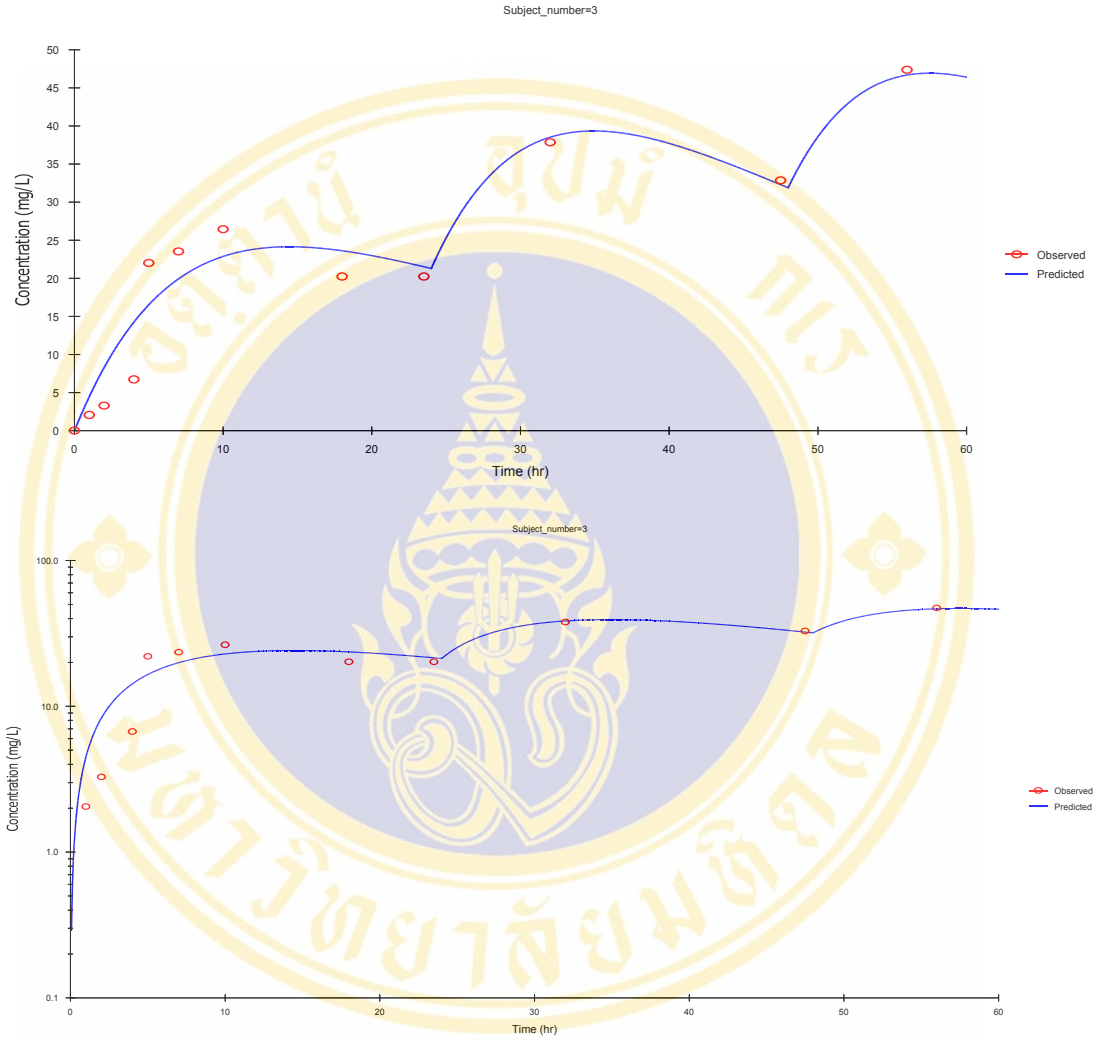

Figure 36 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD06.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

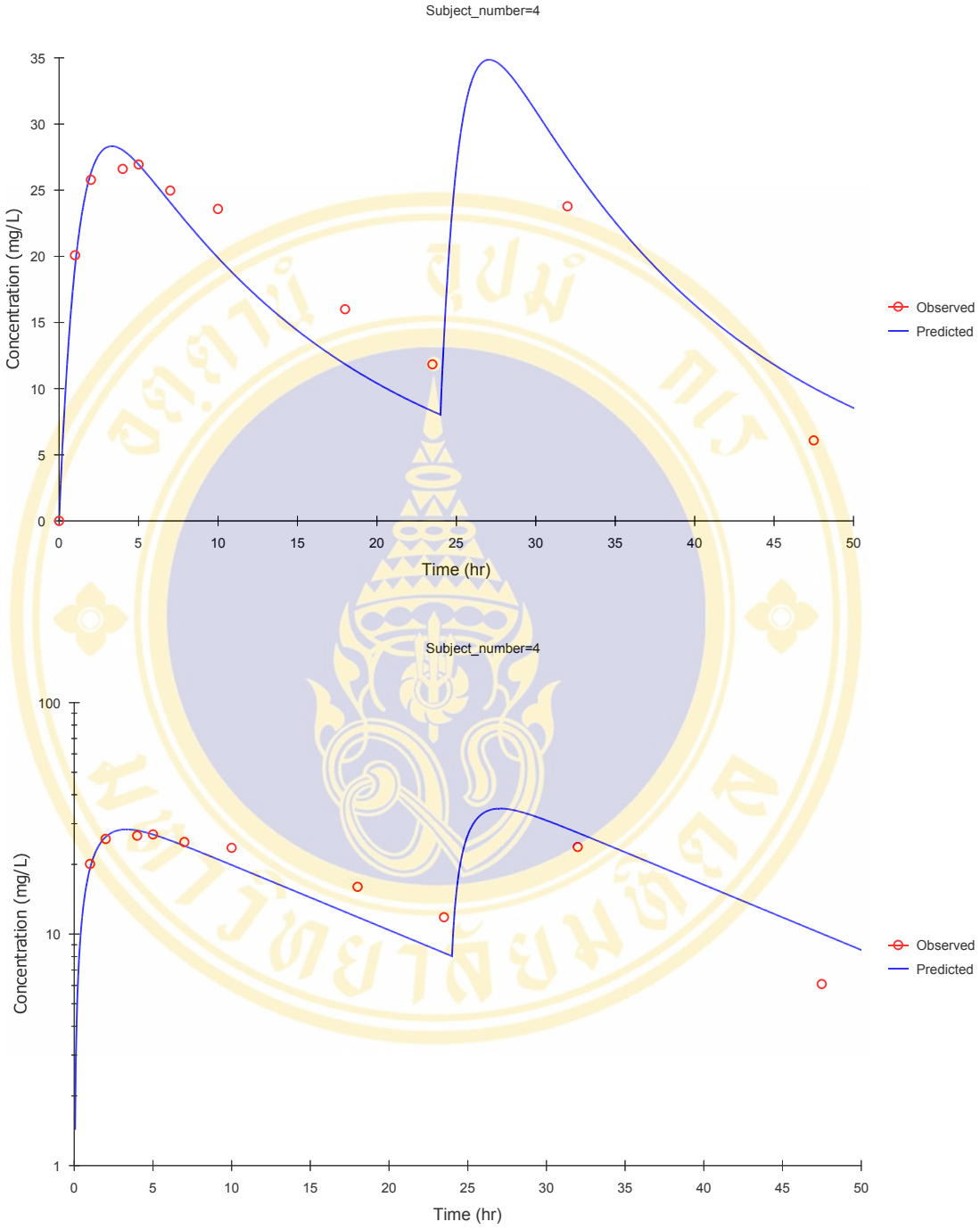

Figure 37 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD08.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

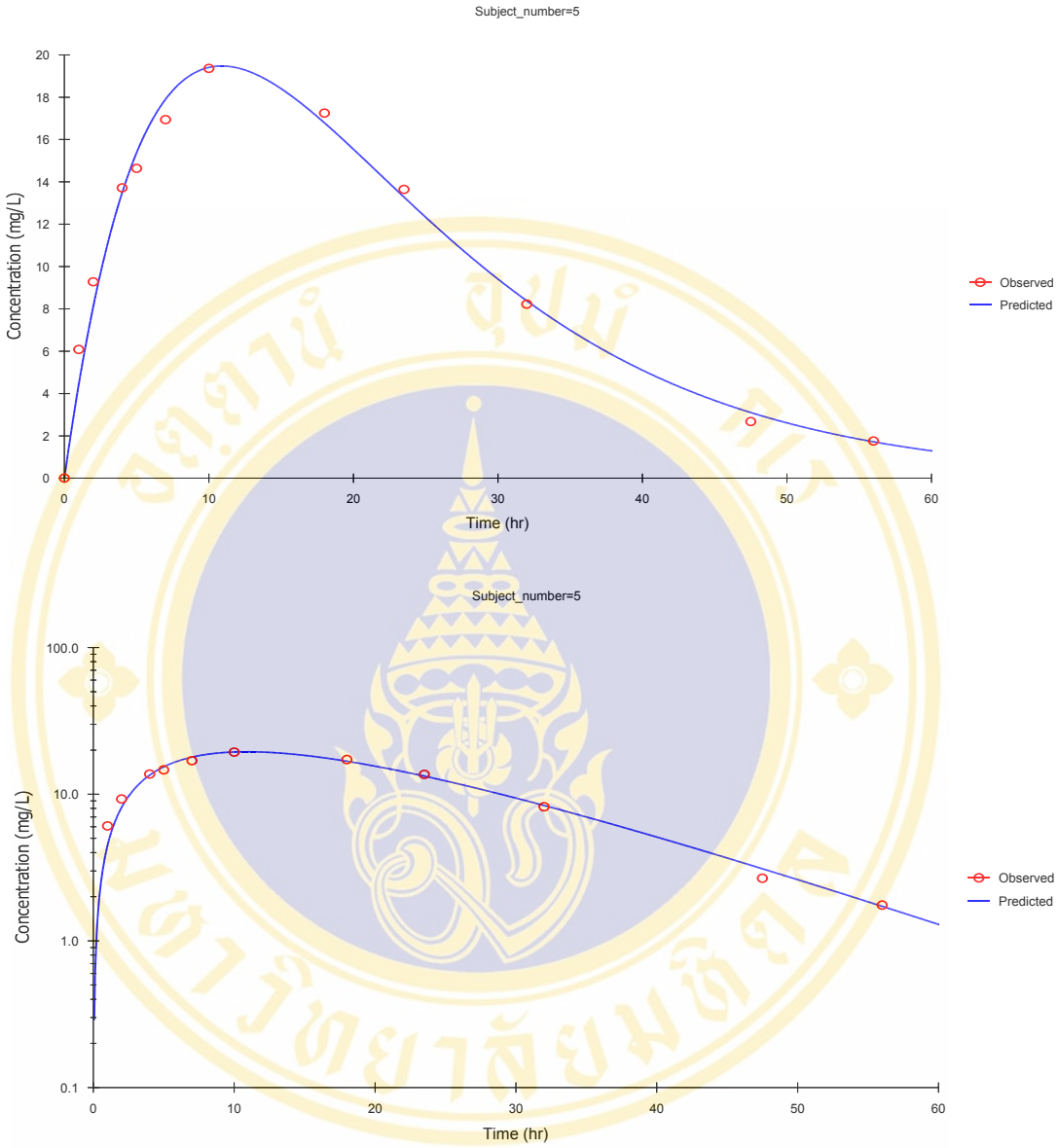

Figure 38 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD10.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

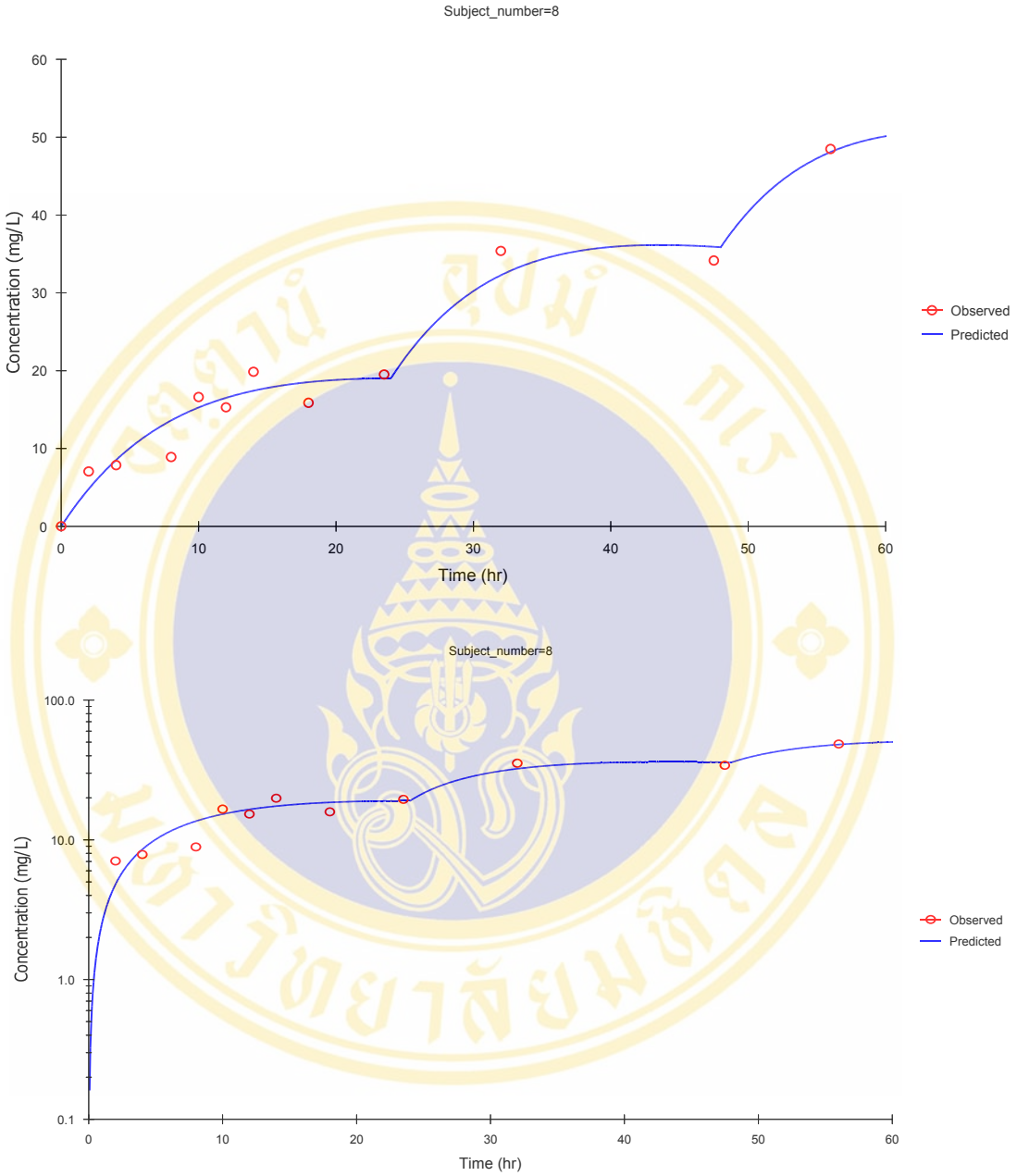

Figure 39 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD15.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

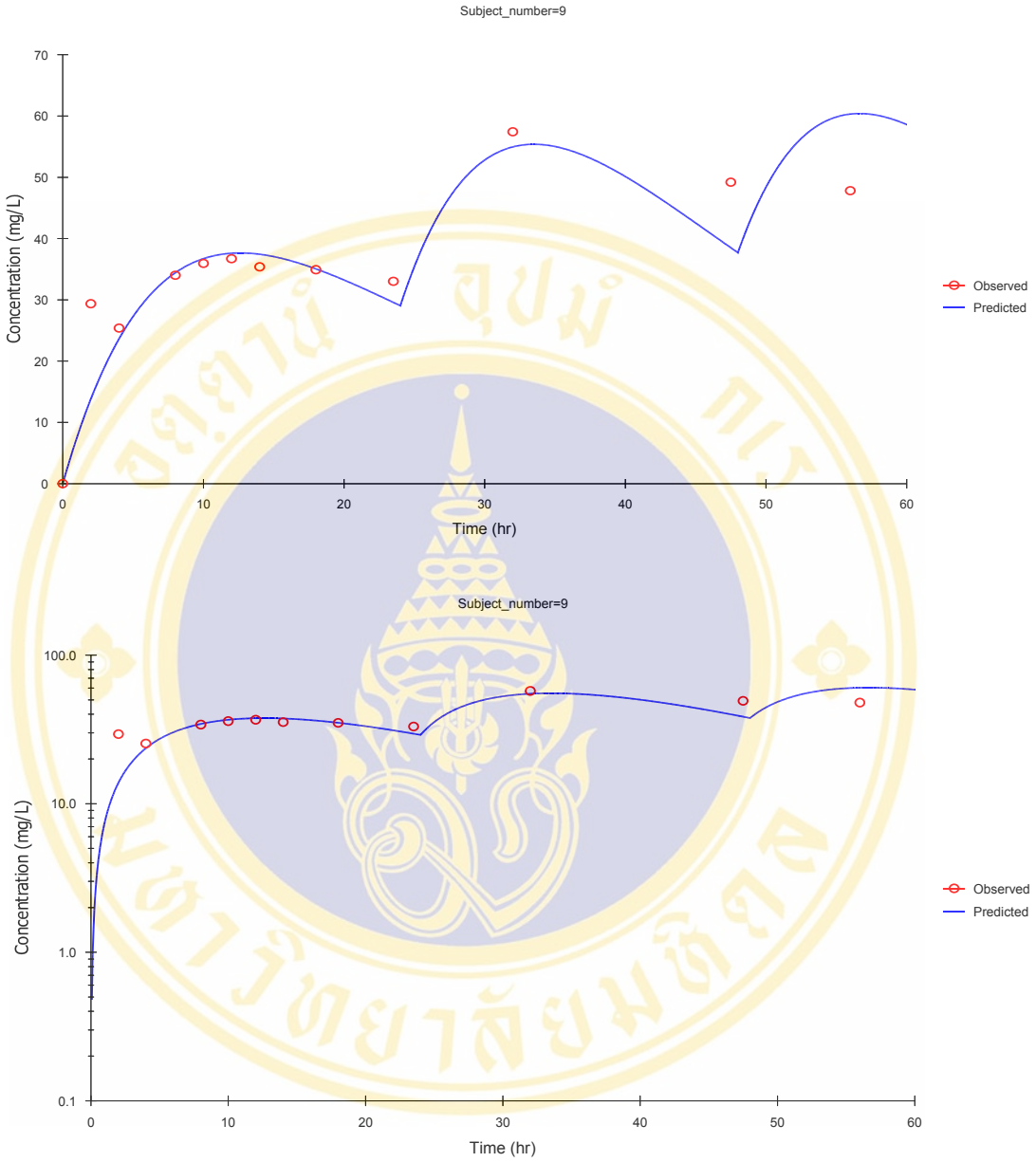

Figure 40 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD18.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

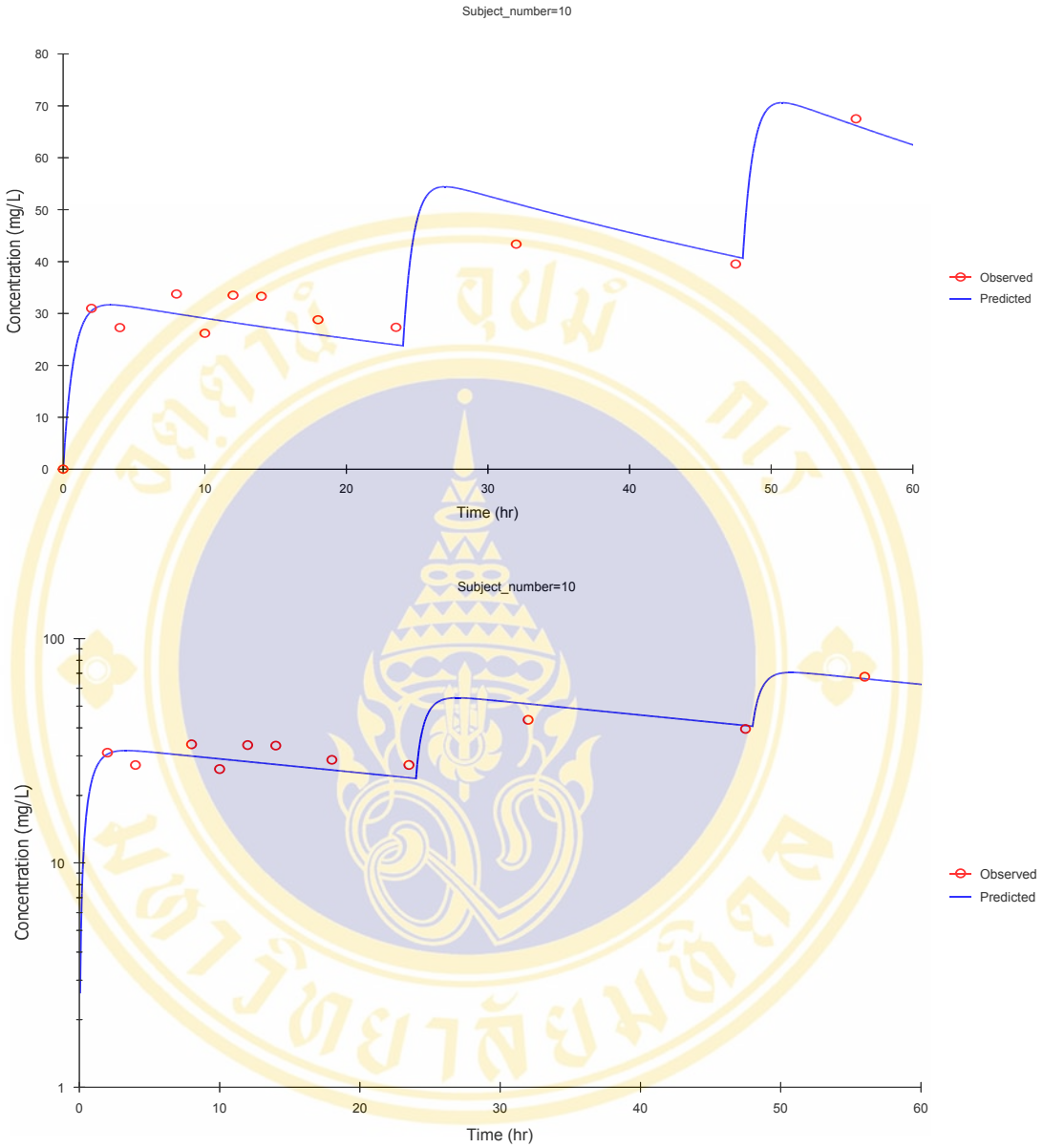

Figure 41 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD19.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

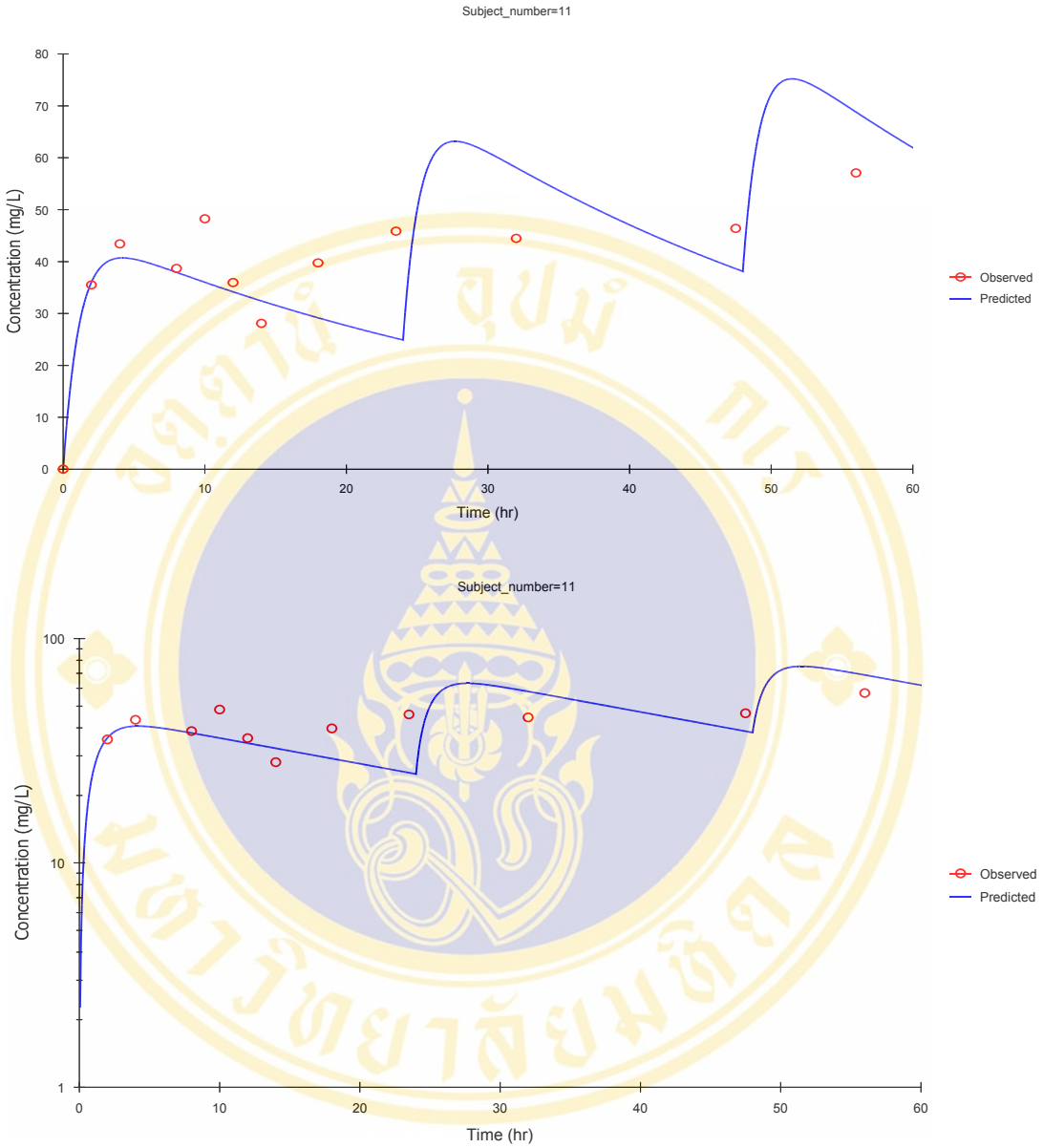

Figure 42 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD21.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

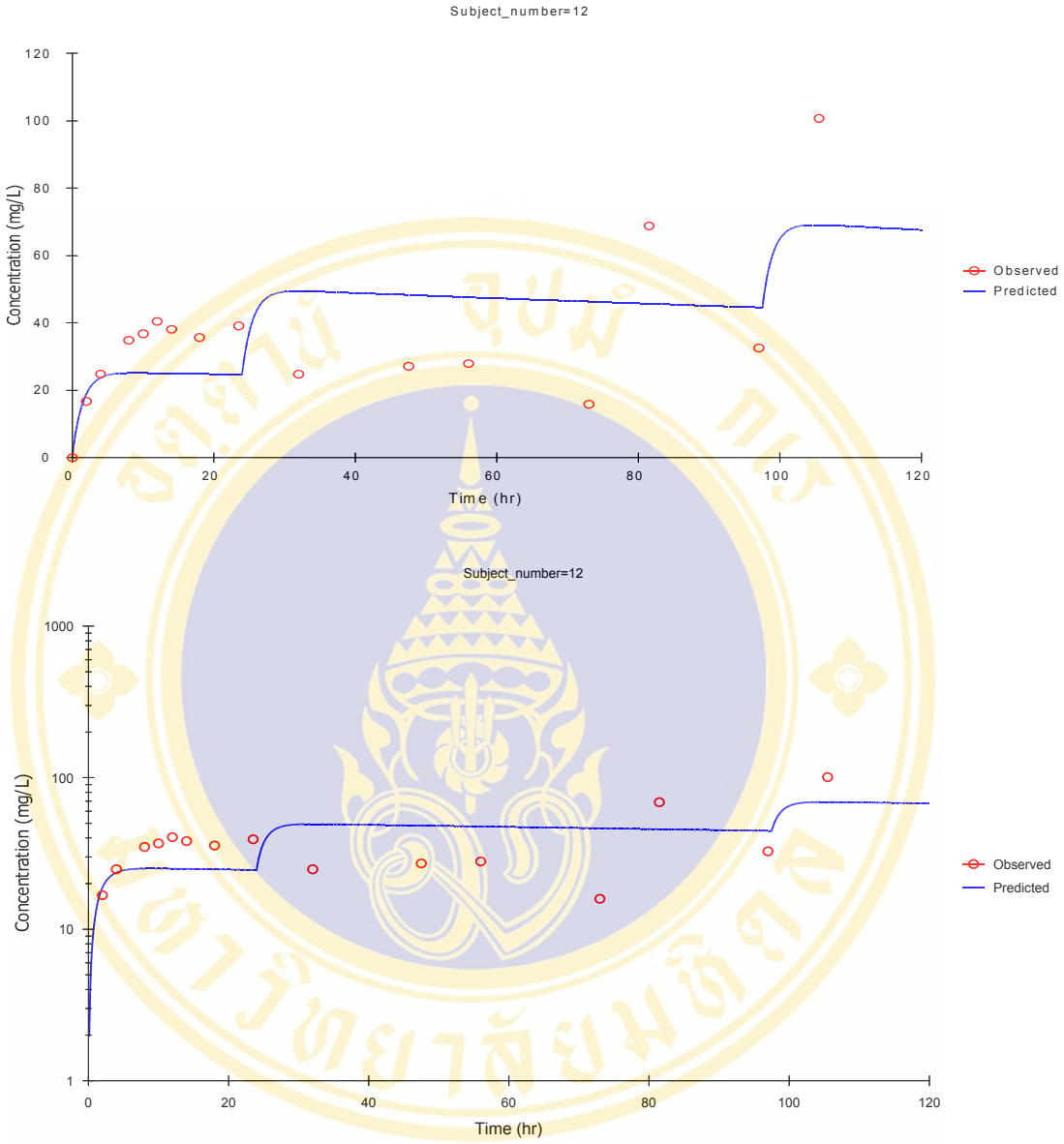

Figure 43 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD24.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

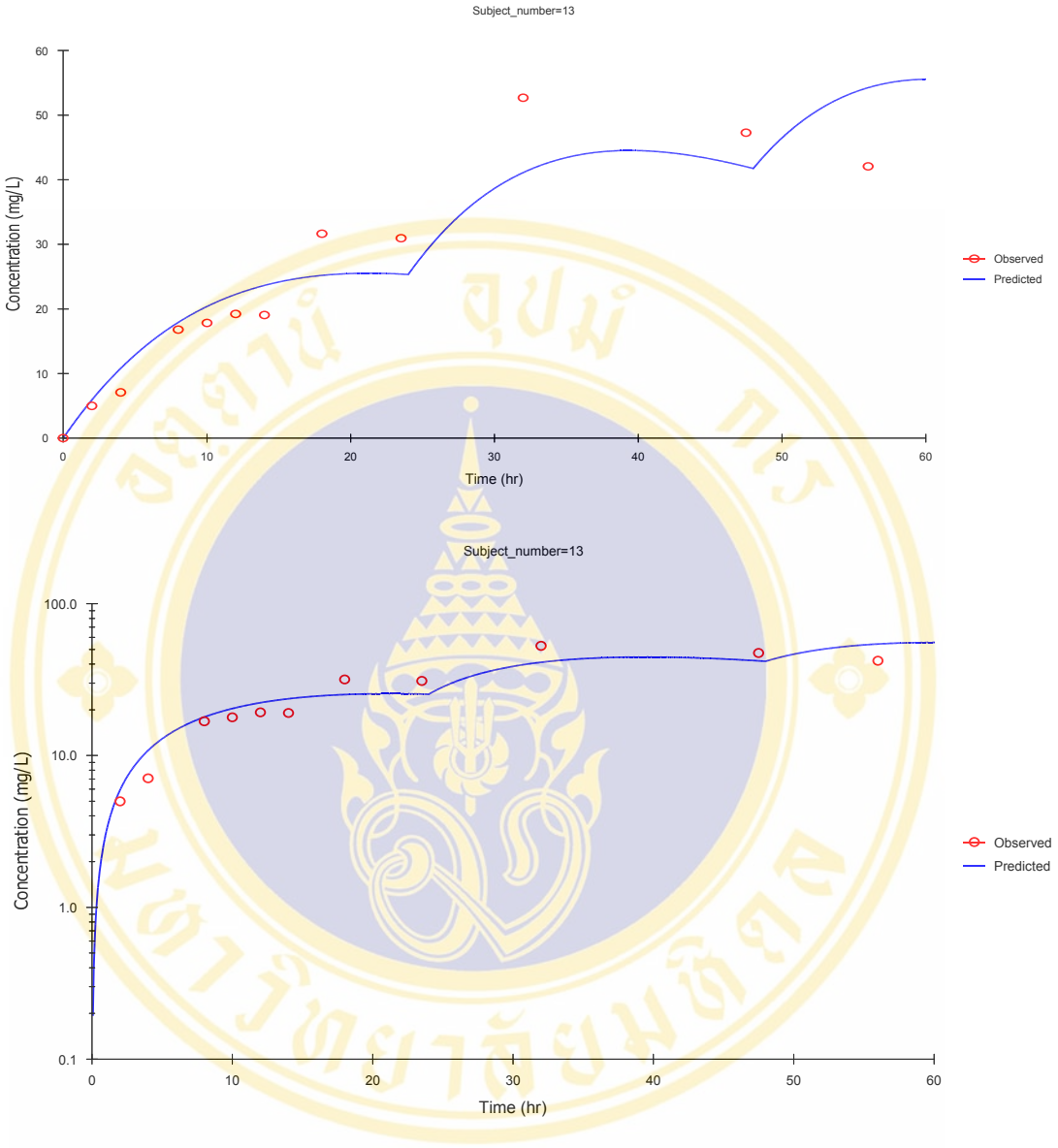

Figure 44 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD25.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

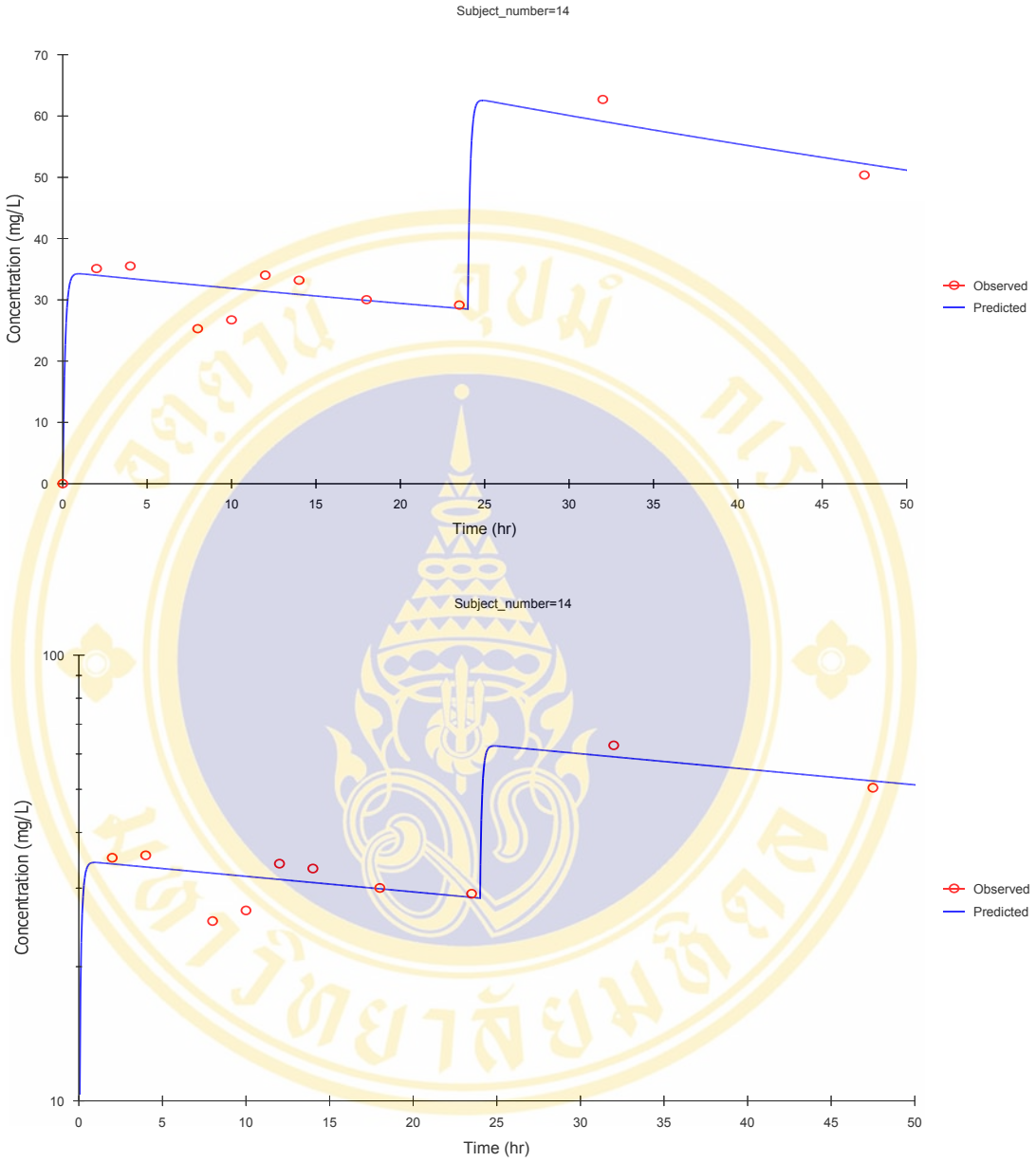

Figure 45 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD27.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

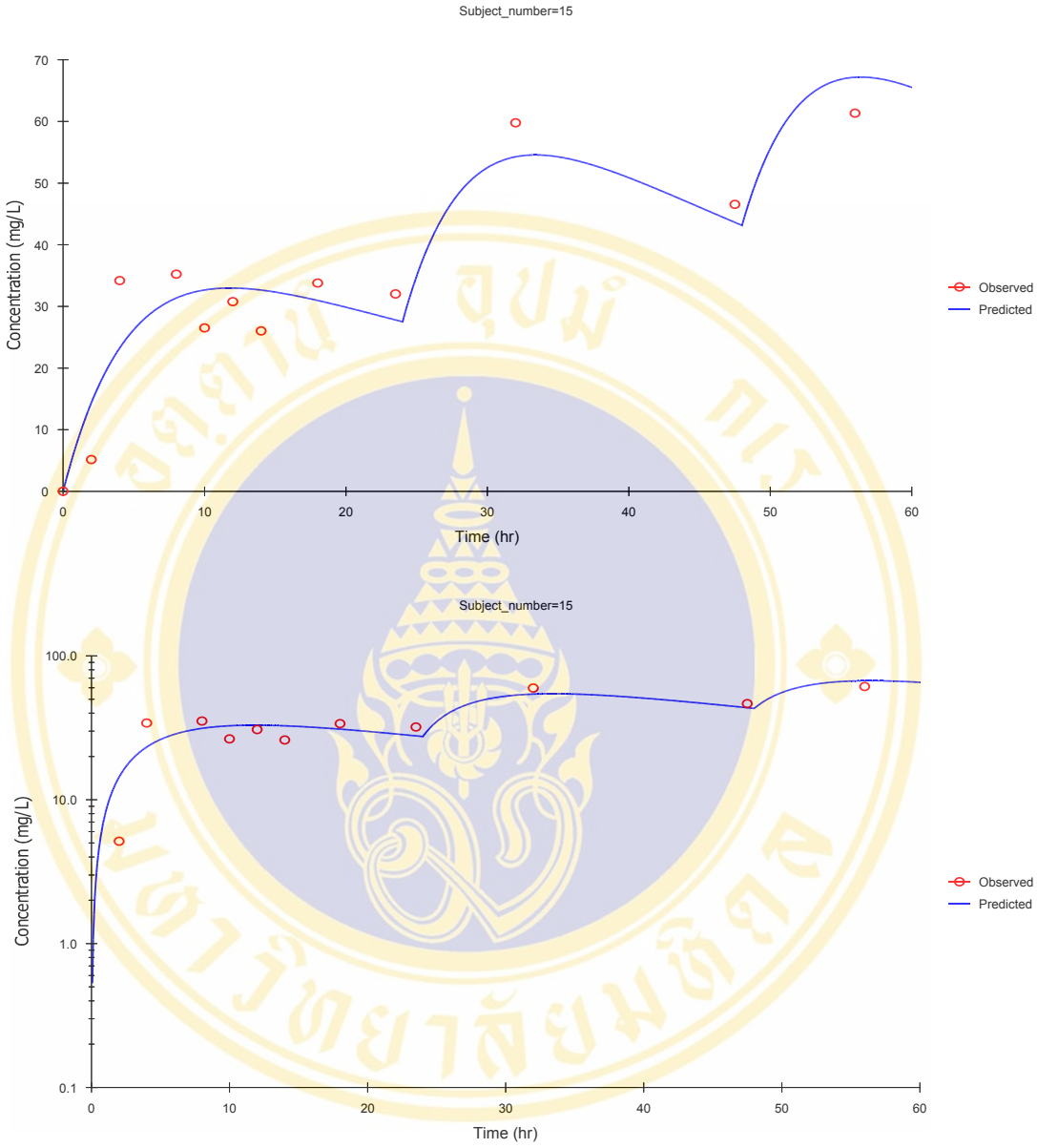

Figure 46 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD30.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

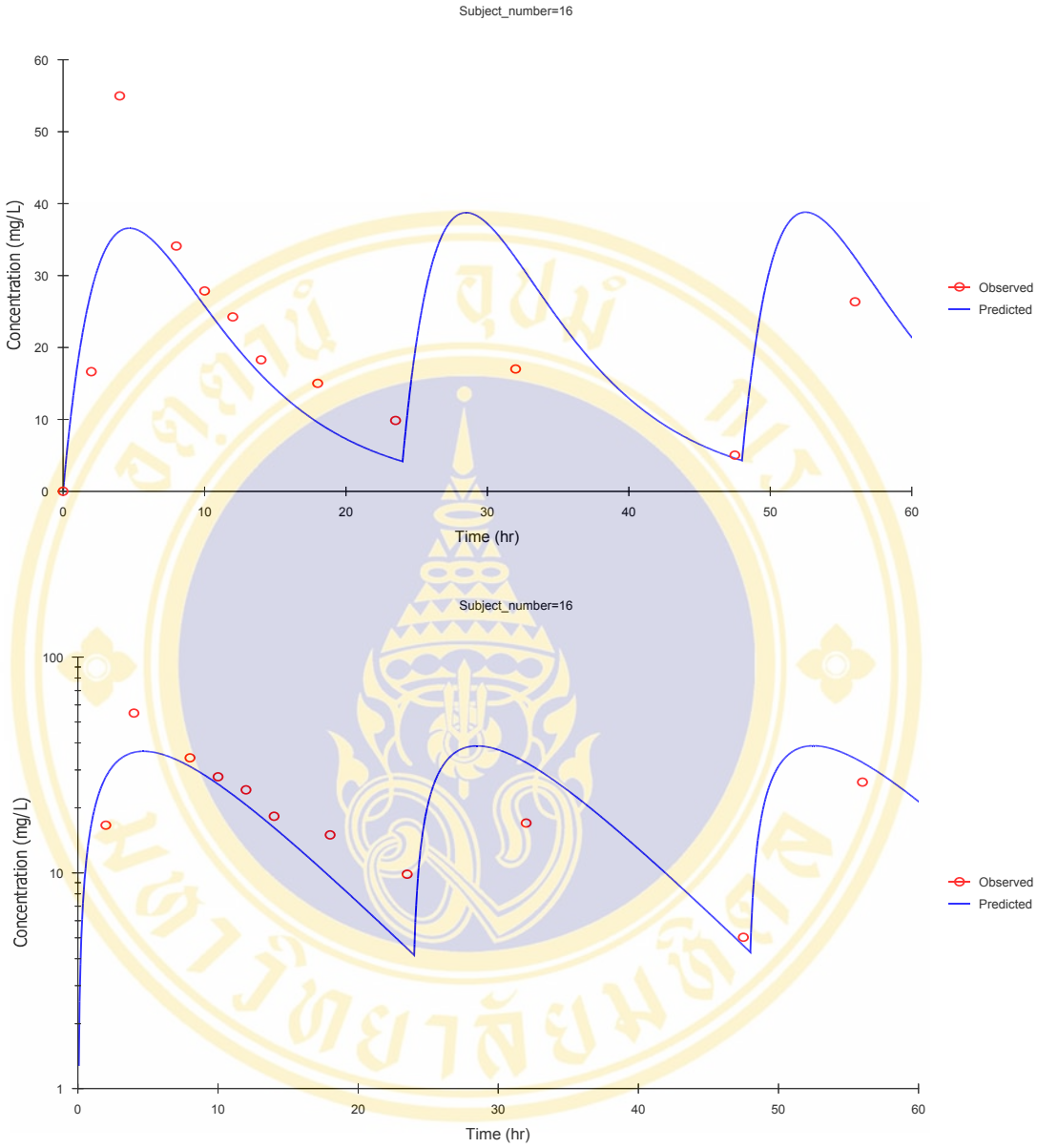

Figure 47 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD32.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

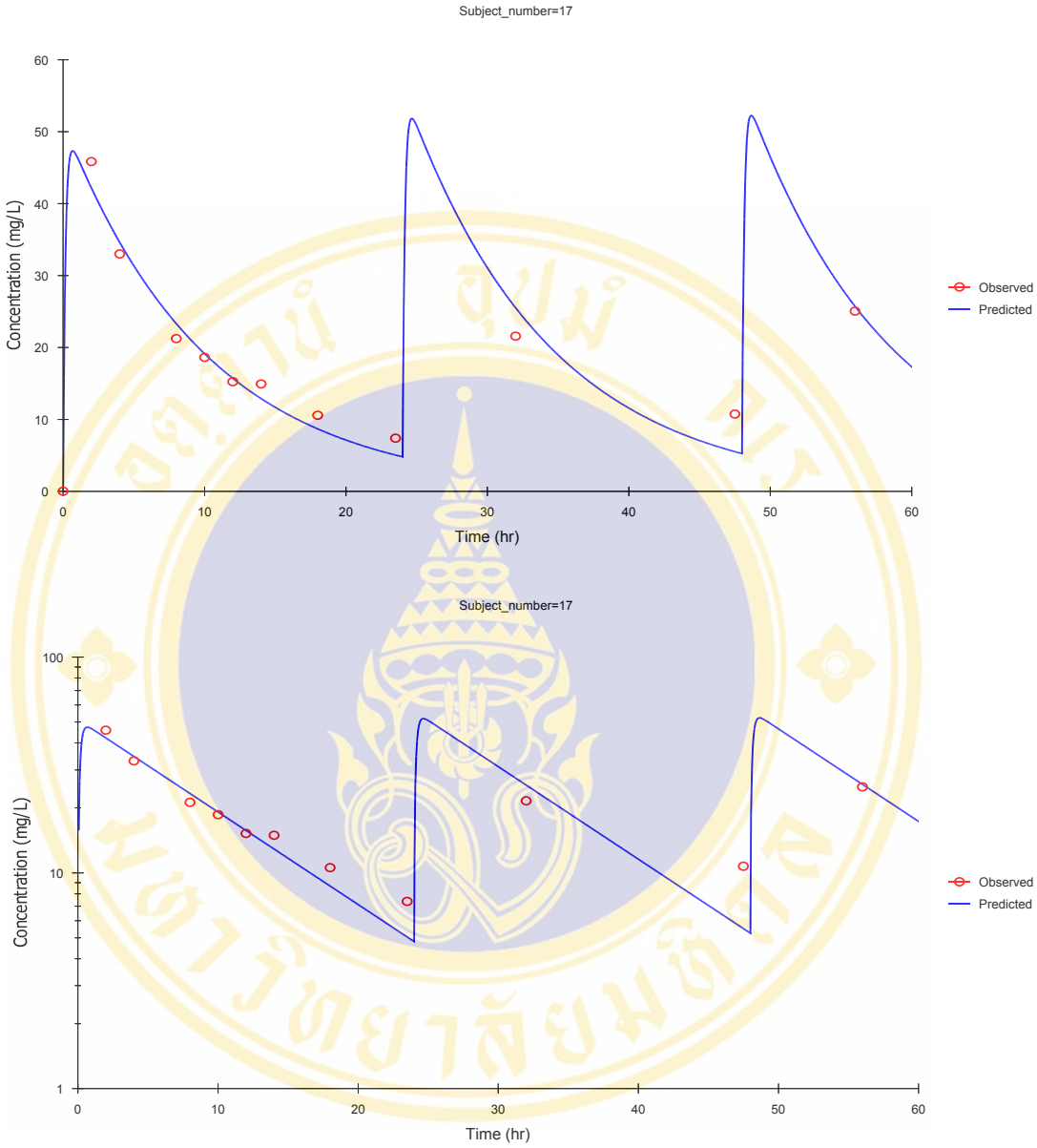

Figure 48 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD34.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

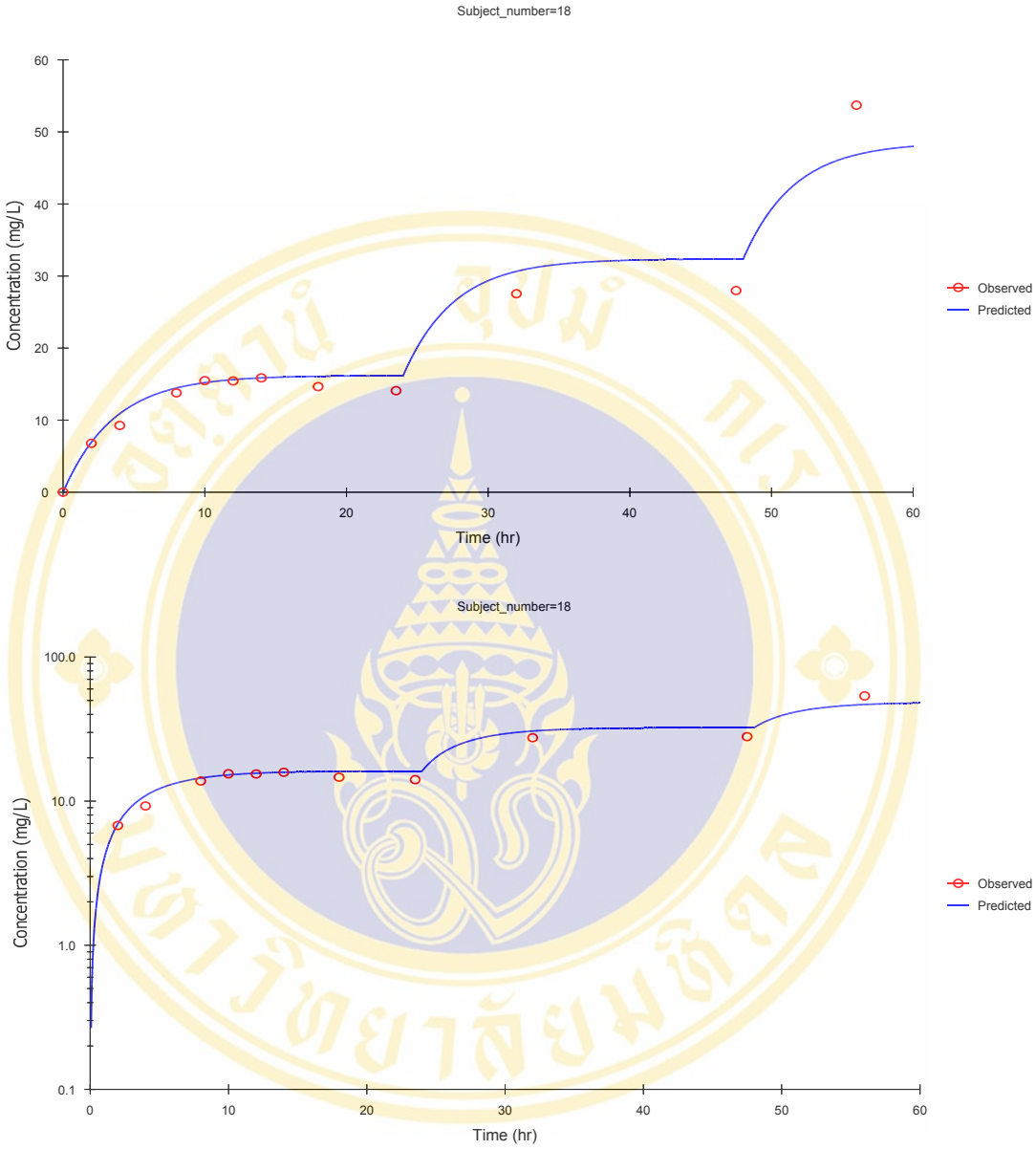

Figure 49 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD36.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

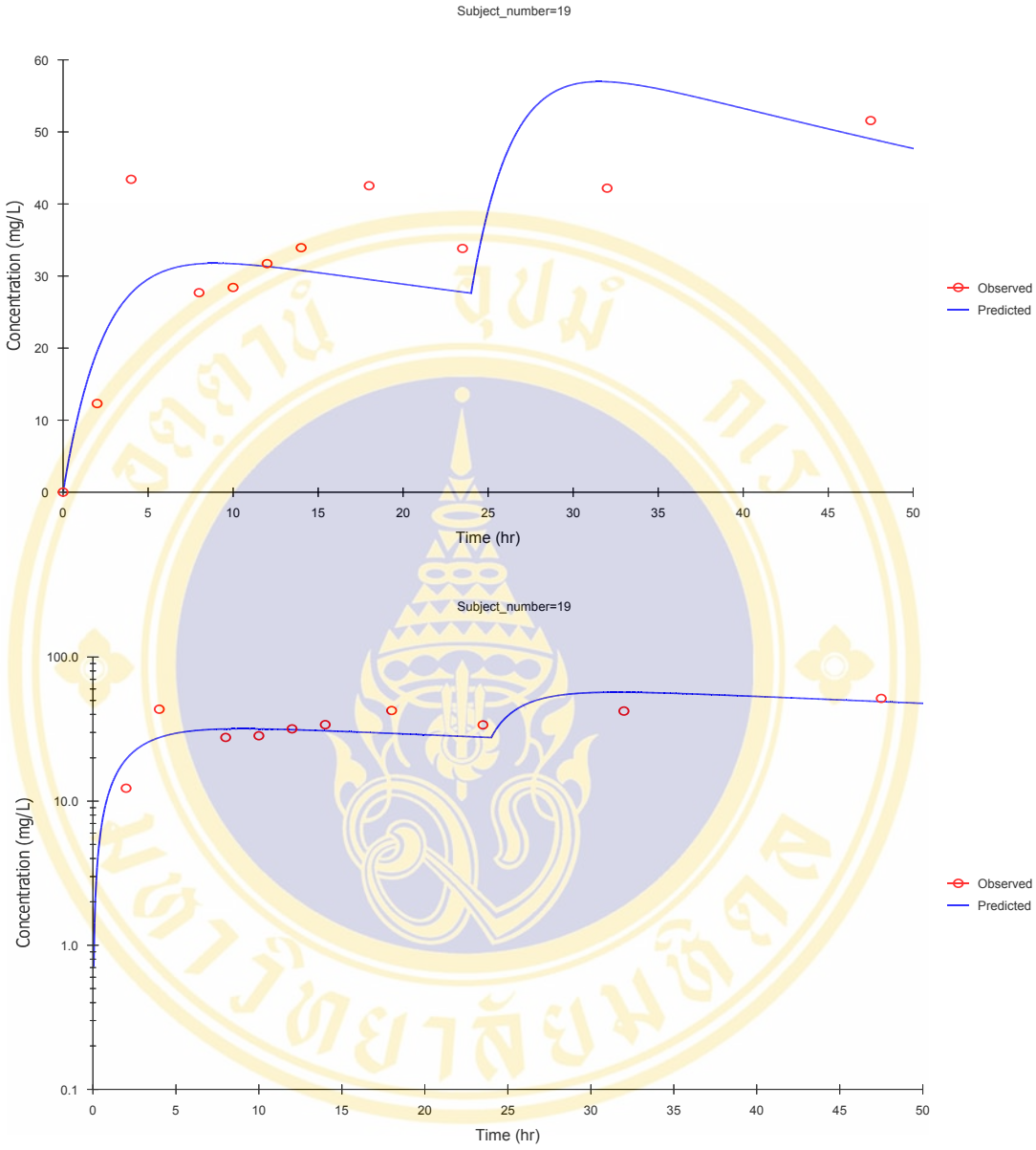

Figure 50 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD38.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

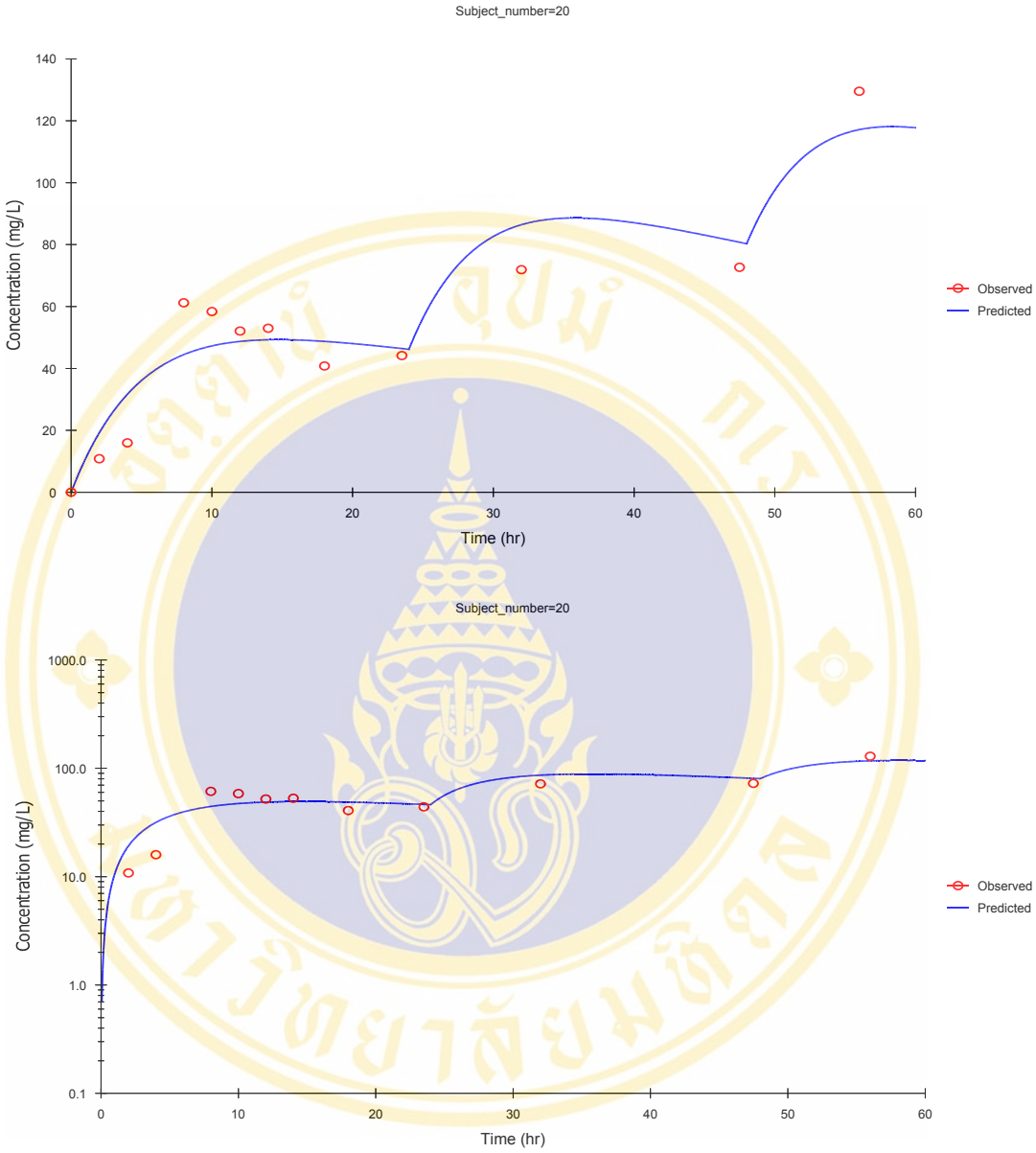

Figure 51 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD39.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

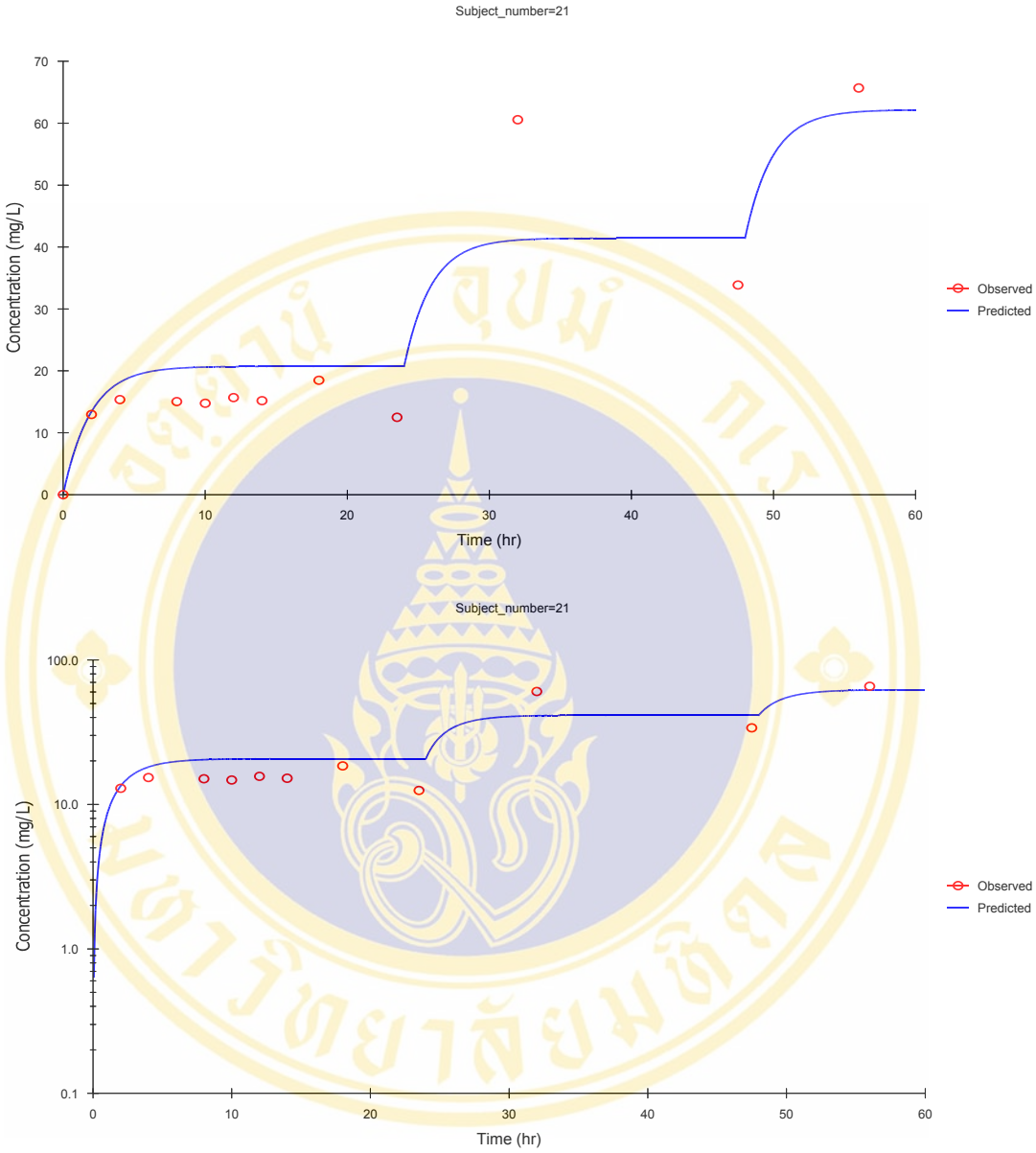

Figure 52 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD42.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

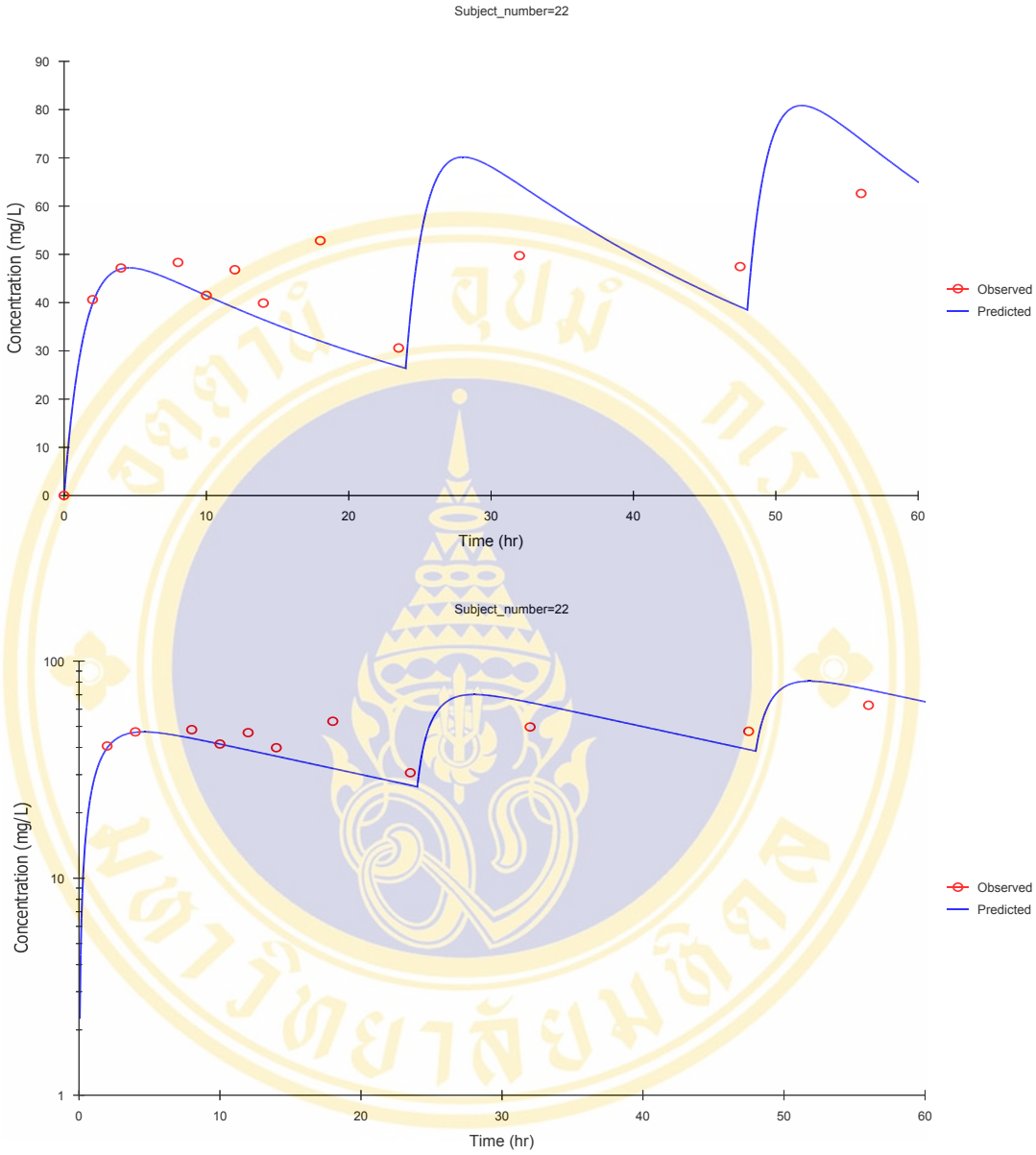

Figure 53 Comparison of plasma ibuprofen concentration between the observed and the predicted values of the subject number RD44.
Upper; Concentration (mg/l) vs time (hr) relation.
Lower; Log concentration (mg/l) vs time (hr) relation.

Clinical record form in control and ibuprofen groups.

Clinical Record Form (A)
Oral ibuprofen suspension administration to prevent symptomatic patent ductus arteriosus in premature infant

Name _____Patient code No._____Drug code No.____
HN_____AN_____ Sex □ Male □ Female
Birth date _____
Gestational age_____weeks by date. Gestational age_____weeks by Ballard score
Birth weight _____gm. Length _____cm. Head circumference_____cm.
Size for date: □ AGA □ SGA □ LGA
Apgar score :1 min _____ 5 min_____ 10 min _____
Mode of delivery: □ spontaneous □ vaginal □ breech □ F/E □ V/E □ C/S
Antenatal steroids □ no □ yes
Respiratory status at entry study:
Ventilated neonates □ no □ yes
Surfactant therapy □ no □ yes
Severity of RDS
OI (Oxygen index = MAP x FiO2 x 100/PaO2) : initial ____heighest ____
VI (Ventilator index = OI x MR) : initial ____heighest ____
Date of administration drugs _____
Age of administration drugs _____hours
PDA evaluation:
□ closed DA at 72 hr
□ back up treatment. Post natal age of treatment _____days
□ surgical ligation
Respiratory out come:
FiO_2 – Day 1 ____ % MAP - Day1 ____ cm H_2O OI ____ VI ____
FiO_2 – Day 2 ____ % MAP - Day2 ____ cm H_2O OI ____ VI ____
FiO_2 – Day 3 ____ % MAP - Day3 ____ cm H_2O OI ____ VI ____
PPHN □ no □ yes
Brochopulmonary dysplasia □ no □ yes stage ____
Days of mechanical ventilation ____
Days of oxygen therapy ____

Fluid intake and renal function evaluation

Fluid intake/ renal function	*Day 0*	*Day 1*	*Day 2*	*Day 3*	*Day 4*	*Day 5*	*Day 6*	*Day 7*
Total fluid intake ml/24 hr	♥	♥	♥	♥	♥	♥	♥	♥
Urine out put(ml/kg/hr)	♥	♥	♥	♥	♥	♥	♥	♥
Serum BUN (mg/dl)	♥	♥	♥	♥				♥
Serum creatinine (mg/dl)	♥	♥	♥	♥				♥
CrCl (ml/min/1.73/m^2)	♥	♥	♥	♥				♥
FENa (%)	♥	♥	♥	♥				♥

Gastrointestinal function and feeding evaluation

Abdominal distension □ no □ yes
Feeding difficulties □ no □ yes
NEC □ no □ yes stage ____
Start of feeding (postnatal age) ____ days
Full oral feeding ____ days

Bleeding disorder

□ no □ yes define _____

Intraventricular hemorrhage (IVH)

□ no □ grade I □ grade II □ grade III □ grade IV

Retinopathy of prematurity (ROP)

□ no □ stage I □ stage II □ stage III □ stage IV

Discharge date _____

Discharge status: □ alive □death

Hospital stay ____ days

Guideline of fluid intake for preterm infant

Date							
Days after admission	**0-1**	**2**	**3**	**4**	**5**	**6**	**7 →**
จำนวน fluid ที่ได้รับ (ml) - ทาง oral - ทาง parenteral รวม ปริมาณที่แนะนำ (ml/kg/day) เด็กคนนี้ควรได้ (ml/day)	60-70	70-80	80-90	90-100	100-110	110-120	120-150
ปริมาณปัสสาวะ (ml) - จำนวนครั้ง - ปริมาณรวม							

การประเมินPDAโดยอาการทางคลินิก

Clinical presentations	D0	D1	D2	D3	D4	D5	D6	D7	D14	D28	หมายเหตุ
Hyperactive precordium											
Bounding pulse (pulse pressure >35 mmHg)											
Continuous systolic murmur											
Cardiomegaly (cardiothoracic ratio > 0.6) or increased pulmonary vasculature											
Hepatomegaly											
Tachycardia(HR>170/min)											

การประเมินภาวะPDA โดย echocardiogram

PDA response	PDA size (mm)	LAE (+ / -)	LVE (+ / -)	Estimated PAP (mmHg)	หมายเหตุ
ก่อนได้รับยา dose แรก (Day 0) วันที่_____ หลังได้รับยา 3 วัน (Day 3) วันที่_____ หลังได้รับยาครบ 7 วัน (Day 7) วันที่_____ หลังได้รับยาครบ 14 วัน (Day 14) วันที่_____ หลังได้รับยาครบ 21 วัน (Day21) วันที่_____ หลังได้รับยาครบ 28วัน (Day 28) วันที่_____					

LAE = Left Atrial Enlargement
LVE = Left Ventricle Enlargement
PAP = Pulmonary Arterial Pressure

Blood chemistry value

Date										
Days after admission	**Normal**	**0**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	หมายเหตุ
Blood Electrolyte		♥			♥					
Na										
K										
Cl										
CO_2										
Renal function		♥			♥					
BUN										
Creatinine										
Clearance										
Coagulation		♥			♥					
Platelet										
PT / INR										
PTT										
CBC		♥			♥					
Hb / Hct										
RBC										
WBC										

ตารางการให้ยาตามน้ำหนักทารก (10mg/kg/dose)

ทารกน้ำหนักตัว_____kg ทารกคนนี้ต้องได้ยาตามรหัสที่ได้รับ ____ mg/dose

การคำนวณคิดจากibuprofen suspension 1 ช้อนชา = 100mg (1ml = 20mg)

ใช้insulin syringeดูดยาโดย 100 ขีด (1ml) = 20mg ดังนั้น 1mg = 5 ขีด

Body weight (gm)	Dose (mg) ที่ได้รับ	ปริมาณยา (ขีดของinsulin syringe)
800	8	40
850	8.5	43
900	9	45
950	9.5	48
1000	10	50
1050	10.5	53
1100	11	55
1150	11.5	58
1200	12	60
1250	12.5	63
1300	13	65
1350	13.5	68
1400	14	70
1450	14.5	73
1500	15	75

ปัญหาที่อาจพบได้ในระหว่างการให้ยาและแนวทางแก้ไข

B ถ้าผู้ป่วยอาเจียนภายใน 45 นาทีหลังได้รับยา dose แรกไปแล้วจะหยุดการให้ยาไปก่อน จนกว่าผู้ป่วยสามารถรับยาได้ตามปกติ หากผู้ป่วยอาเจียนหลังได้รับยา dose แรกไปแล้วเกินกว่า 45 นาที ไม่มีการเปลี่ยนแปลงตารางการให้ยา dose ต่อไปถ้าผู้ป่วยสามารถรับยาได้ตามปกติ

Bถ้าผู้ป่วยเกิดผลข้างเคียงที่รุนแรงจากยาหรือเกิดอาการไม่พึงประสงค์จากยาที่ไม่เคยมีรายงานมาก่อน จะหยุดทำการศึกษาในผู้ป่วยคนดังกล่าวทันทีพร้อมกับให้การรักษาที่เหมาะสมต่อไป

B ถ้าผู้ป่วยเกิดภาวะ transient oliguria (urine output น้อยกว่า 1 ซีซี/น้ำหนักตัว 1 ก.ก./ชั่วโมง) ถ้าค่า BUN ≥ 30 หรือ serum creatinine ≥ 1.5 หลังจากได้รับยา dose แรกหรือ dose ที่สองจะหยุดการศึกษาและให้การรักษาที่เหมาะสมต่อไป

B ถ้าผู้ป่วยเกิดภาวะ NEC (รับอาหารได้น้อยลง เพิ่มpregavage residual อาเจียน abdominal distension พบstool occult bloodในอุจจาระ) จะหยุดทำการศึกษาในผู้ป่วยคนดังกล่าวทันทีพร้อมกับให้การรักษาที่เหมาะสมต่อไป

Clinical Record Form (B)

Oral ibuprofen suspension administration to prevent symptomatic patent ductus arteriosus in premature infant

Name _____Patient code No._____Drug code No.____
HN_____AN_____ Sex □ Male □ Female
Birth date _____
Gestational age_____weeks by date. Gestational age_____weeks by Ballard score
Birth weight _____gm. Length _____cm. Head circumference_____cm.
Size for date: □ AGA □ SGA □ LGA
Apgar score :1 min _____ 5 min_____ 10 min _____
Mode of delivery: □ spontaneous □ vaginal □ breech □ F/E □ V/E □ C/S
Antenatal steroids □ no □ yes
Respiratory status at entry study:
Ventilated neonates □ no □ yes
Surfactant therapy □ no □ yes
Severity of RDS
OI (Oxygen index = MAP x FiO2 x 100/PaO2) : initial ____heighest ____
VI (Ventilator index = OI x MR) : initial ____heighest ____
Date of administration drugs _____
Age of administration drugs _____hours
PDA evaluation:
□ closed DA at 72 hr
□ back up treatment. Post natal age of treatment _____days
□ surgical ligation
Respiratory out come:
FiO_2 – Day 1 ____ % MAP - Day1 ____ cm H_2O OI ____ VI ____
FiO_2 – Day 2 ____ % MAP - Day2 ____ cm H_2O OI ____ VI ____
FiO_2 – Day 3 ____ % MAP - Day3 ____ cm H_2O OI ____ VI ____
PPHN □ no □ yes
Brochopulmonary dysplasia □ no □ yes stage ____
Days of mechanical ventilation ____
Days of oxygen therapy ____

Fluid intake and renal function evaluation

Fluid intake/ renal function	*Day 0*	*Day 1*	*Day 2*	*Day 3*	*Day 4*	*Day 5*	*Day 6*	*Day 7*
Total fluid intake ml/24 hr	♥	♥	♥	♥	♥	♥	♥	♥
Urine out put(ml/kg/hr)	♥	♥	♥	♥	♥	♥	♥	♥
Serum BUN (mg/dl)	♥	♥	♥	♥				♥
Serum creatinine (mg/dl)	♥	♥	♥	♥				♥
CrCl (ml/min/1.73/m^2)	♥	♥	♥	♥				♥
FENa (%)	♥	♥	♥	♥				♥

Gastrointestinal function and feeding evaluation

Abdominal distension □ no □ yes
Feeding difficulties □ no □ yes
NEC □ no □ yes stage ____
Start of feeding (postnatal age) ____ days
Full oral feeding ____ days

Bleeding disorder

□ no □ yes define _____

Intraventricular hemorrhage (IVH)

□ no □ grade I □ grade II □ grade III □ grade IV

Retinopathy of prematurity (ROP)

□ no □ stage I □ stage II □ stage III □ stage IV

Discharge date _____

Discharge status: □ alive □death

Hospital stay ____ days

Guideline of fluid intake for preterm infant

Date							
Days after admission	**0-1**	**2**	**3**	**4**	**5**	**6**	**7 →**
จำนวน fluid ที่ได้รับ (ml) - ทาง oral - ทาง parenteral รวม ปริมาณที่แนะนำ (ml/kg/day) เด็กคนนี้ควรได้ (ml/day)	60-70	70-80	80-90	90-100	100-110	110-120	120-150
ปริมาณปัสสาวะ (ml) - จำนวนครั้ง - ปริมาณรวม							

การประเมินPDAโดยอาการทางคลินิก

Clinical presentations	D0	D1	D2	D3	D4	D5	D6	D7	D14	D28	หมายเหตุ
Hyperactive precordium											
Bounding pulse (pulse pressure >35 mmHg)											
Continuous systolic murmur											
Cardiomegaly (cardiothoracic ratio > 0.6) or increased pulmonary vasculature											
Hepatomegaly											
Tachycardia (HR>170/min)											

การประเมินภาวะPDA โดย echocardiogram

PDA response	PDA size (mm)	LAE (+ / -)	LVE (+ / -)	Estimated PAP (mmHg)	หมายเหตุ
ก่อนได้รับยา dose แรก (Day 0) วันที่_____ หลังได้รับยา 3 วัน (Day 3) วันที่_____ หลังได้รับยาครบ 7 วัน (Day 7) วันที่_____ หลังได้รับยาครบ 14 วัน(Day 14) วันที่_____ หลังได้รับยาครบ21วัน (Day21) วันที่_____ หลังได้รับยาครบ 28วัน (Day 28) วันที่_____					

LAE = Left Atrial Enlargement
LVE = Left Ventricle Enlargement
PAP = Pulmonary Arterial Pressure

Blood chemistry value

Date										
Days after admission	**Normal**	**0**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	หมายเหตุ
Blood Electrolyte		♥			♥					
Na										
K										
Cl										
Co_2										
Renal function		♥			♥					
BUN										
Creatinine										
Clearance										
Coagulation		♥			♥					
Platelet										
PT / INR										
PTT										
CBC		♥			♥					
Hb / Hct										
RBC										
WBC										

ตารางการให้ยาตามน้ำหนักทารก (10mg/kg/dose)

ทารกน้ำหนักตัว_____kg ทารกคนนี้ต้องได้ยาตามรหัสที่ได้รับ ____ mg/dose

การคำนวณคิดจากibuprofen suspension 1 ช้อนชา = 100mg (1ml = 20mg)

ใช้insulin syringeดูดยาโดย 100 ขีด (1ml) = 20mg ดังนั้น 1mg = 5 ขีด

Body weight (gm)	Dose (mg) ที่ได้รับ	ปริมาณยา (ขีดของinsulin syringe)
800	8.0	40
850	8.5	43
900	9.0	45
950	9.5	48
1000	10.0	50
1050	10.5	53
1100	11.0	55
1150	11.5	58
1200	12.0	60
1250	12.5	63
1300	13.0	65
1350	13.5	68
1400	14.0	70
1450	14.5	73
1500	15.0	75

ปัญหาที่อาจพบได้ในระหว่างการให้ยาและแนวทางแก้ไข

B ถ้าผู้ป่วยอาเจียนภายใน 45 นาทีหลังได้รับยา dose แรกไปแล้วจะหยุดการให้ยาไปก่อน จนกว่าผู้ป่วยสามารถรับยาได้ตามปกติ หากผู้ป่วยอาเจียนหลังได้รับยา dose แรกไปแล้วเกิน กว่า 45 นาที ไม่มีการเปลี่ยนแปลงตารางการให้ยา dose ต่อไปถ้าผู้ป่วยสามารถรับยาได้ตามปกติ

Bถ้าผู้ป่วยเกิดผลข้างเคียงที่รุนแรงจากยาหรือเกิดอาการไม่พึงประสงค์จากยาที่ไม่เคยมีรายงานมาก่อน จะหยุดทำการศึกษาในผู้ป่วยคนดังกล่าวทันทีพร้อมกับให้การรักษาที่เหมาะสมต่อไป

B ถ้าผู้ป่วยเกิดภาวะ transient oliguria (urine output น้อยกว่า 1 ซีซี/น้ำหนักตัว 1 ก.ก./ชั่วโมง) ถ้าค่า BUN ≥ 30 หรือ serum creatinine ≥ 1.5 หลังจากได้รับยา dose แรกหรือ dose ที่สองจะหยุดการศึกษาและให้การรักษาที่เหมาะสมต่อไป

B ถ้าผู้ป่วยเกิดภาวะ NEC (รับอาหารได้น้อยลง เพิ่มpregavage residual อาเจียน abdominal distension พบstool occult bloodในอุจจาระ) จะหยุดทำการศึกษาในผู้ป่วยคนดังกล่าวทันทีพร้อมกับให้การรักษาที่เหมาะสมต่อไป

ข้อมูลการเจาะเลือดของผู้ป่วย

คำนวณปริมาณเลือดที่ใช้ทั้งหมด

1. ปริมาณเลือดที่เจาะรวมทั้งหมดไม่เกิน 5-10% total body weight คิดเป็น …….ml
2. ปริมาณเลือดที่เจาะสำหรับเด็กคนนี้

2.1Biochemistry (BUN,Cr,Electrolyte,CBC,Coagulogram)…….ml

2.2 จากการเจาะเลือดที่เวลาต่างๆรวม …… ml

2.3 ปริมาณรวมทั้งหมด …… ml □ เกินปริมาณแนะนำ …… ml □ ไม่เกิน

วันและเวลาที่เจาะเลือด	ปริมาณเลือด (ml)	ลายเซ็น ผู้เจาะเลือด	หมายเหตุ
Dose ที่ 1 วันที่_____			
เวลาที่เริ่มให้ยา เจาะเลือดที่เวลา_____			
2 ชั่วโมงหลังได้รับยา เจาะเลือดที่เวลา_____			
4 ชั่วโมงหลังได้รับยา เจาะเลือดที่เวลา_____			
8 ชั่วโมงหลังได้รับยา เจาะเลือดที่เวลา_____			
10 ชั่วโมงหลังได้รับยา เจาะเลือดที่เวลา_____			
12 ชั่วโมงหลังได้รับยา เจาะเลือดที่เวลา_____			
14 ชั่วโมงหลังได้รับยา เจาะเลือดที่เวลา_____			
18 ชั่วโมงหลังได้รับยา เจาะเลือดที่เวลา_____			
Dose ที่ 2 วันที่_____			
30 นาที ก่อนได้รับยา เจาะเลือดที่เวลา_____			
เวลาที่เริ่มให้ยา _____			
8 ชั่วโมงหลังได้รับยา เจาะเลือดที่เวลา_____			
Dose ที่ 3 วันที่_____			
30 นาที ก่อนได้รับยา เจาะเลือดที่เวลา_____			
เวลาที่เริ่มให้ยา _____			
8 ชั่วโมงหลังได้รับยา เจาะเลือดที่เวลา_____			

Informed consent for recruited subjects.

โครงการศึกษาวิจัย

เรื่อง

การใช้ยา Ibuprofen ชนิดรับประทาน ในการป้องกันการเกิด symptomatic patent ductus arteriosus ในทารกคลอดก่อนกำหนด (Oral ibuprofen suspension administration to prevent symptomatic patent ductus arteriosus in premature infant)

เอกสารแนะนำสำหรับอาสาสมัคร

บทนำ

ทารกในครรภ์จะมีหลอดเลือดที่เรียกว่า ductus arteriosus เชื่อมต่อระหว่างหลอดเลือดที่ไปปอดและหลอดเลือดแดงใหญ่ ซึ่งจะปิดได้เองภายหลังคลอด ในทารกคลอดครบกำหนดจะปิดได้เร็วกว่าทารกคลอดก่อนกำหนด ถ้าหลอดเลือดนี้ยังคงเปิดอยู่จะทำให้เลือดไหลย้อนไปยังปอดมากขึ้น ทำให้ทารกหายใจลำบาก ต้องใช้เครื่องช่วยหายใจ ต้องการออกซิเจนเพิ่มขึ้นเสี่ยงต่อการเกิดปอดอักเสบเรื้อรังและหัวใจล้มเหลวได้ และอาจมีเลือดออกในสมองได้โดยเฉพาะทารกที่มีอายุครรภ์และน้ำหนักตัวน้อย การรักษาภาวะ PDA แบ่งออกได้เป็นการรักษาแบบประคับประคองและการรักษาแบบจำเพาะโดยการให้ยาหรือผ่าตัด มาตรฐานการรักษาในปัจจุบันจะให้ยาเมื่อมีอาการ ร่วมกับจำกัดปริมาณสารน้ำให้เพียงพอในแต่ละวัน และการรักษาประคับประคองอื่นๆยาที่ใช้รักษาตัวแรกคือ Indomethacin ชนิดฉีด ซึ่งมีประสิทธิภาพสูงในการปิดหลอดเลือด ductus arteriosus แต่มีผลข้างเคียงสูงเช่นกัน ทำให้มีเลือดออกง่าย เลือดไปเลี้ยงไตน้อยลง ปัสสาวะออกน้อย ไตวาย ลำไส้เปื่อยเน่าได้ ผู้ป่วยต้องอยู่โรงพยาบาลนานขึ้น ปัจจุบันมีการใช้ยา Ibuprofen ชนิดฉีด ในการรักษา อาการ PDA ในทารกแรกคลอดก่อนกำหนดพบว่าได้ผลดีเช่นเดียวกับยา Indomethacin ชนิดฉีด แต่มีผลข้างเคียงน้อยกว่า และยังสามารถใช้ปิดหลอดเลือด DA ป้องกันการเกิดอาการ PDA

คณะวิจัยจึงได้ทำการศึกษาครั้งนี้ขึ้นเพื่อประเมินประสิทธิภาพของยา Ibuprofen ในการป้องกันการเกิดอาการ PDA ในทารกแรกคลอดก่อนกำหนด การศึกษาครั้งนี้จะมีการเจาะเลือดทารกและหาค่าทางเภสัชจลนศาสตร์ในการทำนายระดับยาที่เหมาะสมที่จำเป็นต่อการปิดของ ductus arteriosus ได้ และประเมินผลข้างเคียงที่อาจเกิดขึ้นด้วย

โครงการศึกษาวิจัยนี้จะดำเนินการโดยหน่วยทารกแรกเกิด กลุ่มงานกุมารเวชกรรม สถาบันสุขภาพเด็กแห่งชาติมหาราชินี กรมการแพทย์ กระทรวงสาธารณสุข ร่วมกับคณะเภสัชศาสตร์ มหาวิทยาลัยมหิดล โดยจะศึกษาในทารกที่คลอดก่อนกำหนด 42 คนที่เข้ารับการรักษาในสถาบันสุขภาพเด็กแห่งชาติมหาราชินี

วัตถุประสงค์ของโครงการ

1. **เพื่อศึกษาประสิทธิภาพการใช้ยาน้ำ Ibuprofen ชนิดรับประทาน ในการป้องกันการเกิด symptomatic PDA**
2. เพื่อศึกษาค่าทางเภสัชจลนศาสตร์ของยาน้ำ Ibuprofen ชนิดรับประทานในทารกแรกคลอดก่อนกำหนด
3. เพื่อศึกษาผลข้างเคียงจากยาน้ำ Ibuprofen ชนิดรับประทาน Ibuprofen ต่อระบบที่สำคัญของร่างกาย เช่น ไต ระบบทางเดินอาหารและตับ

เนื่องจากทารกของท่านคลอดก่อนกำหนดและมีโอกาสเสี่ยงที่จะเกิดอาการ PDA ท่านและบุตรจึงได้รับเชิญให้เข้าร่วมโครงการศึกษาวิจัย ก่อนที่ท่านจะตัดสินใจอนุญาตให้บุตรของท่านเข้าร่วมในการศึกษานี้ ท่านต้องเข้าใจถึงข้อดีข้อเสียของการเข้าร่วมโครงการ เพื่อท่านจะสามารถตัดสินใจได้โดยท่านมีความรู้ที่ถูกต้องตามความเป็นจริง เราจึงเรียกการให้คำยินยอมแบบนี้ว่าการให้ความยินยอมโดยได้รับความรู้ที่ถูกต้องตามความเป็นจริง

ใบยินยอมฉบับนี้จะให้ข้อมูลเกี่ยวกับการวิจัยซึ่งจะถูกนำมาพูดคุยกับท่าน เมื่อท่านเข้าใจการศึกษา และขั้นตอนต่างๆในการวิจัยแล้ว หากท่านมีความประสงค์ให้บุตรเข้าร่วมการศึกษาท่านจะถูกขอร้องให้ลงลายมือชื่อในใบยินยอมฉบับนี้ การตัดสินใจยินยอมให้บุตรเข้าร่วมในการศึกษาครั้งนี้เป็นไปโดยความสมัครใจ ซึ่งหมายความว่าท่านมีอิสระในการจะให้บุตรเข้าร่วมและมีอิสระที่จะให้บุตรออกจากโครงกาเมื่อใดก็ได้ การตัดสินใจไม่ให้บุตรเข้าร่วมโครงการนี้จะไม่มีผลใดๆต่อการรักษาของบุตร ที่จะได้รับจากแพทย์ผู้วิจัย

ขั้นตอนการดำเนินโครงการศึกษาวิจัย

1. ก่อนเข้าร่วมโครงการแพทย์ผู้วิจัยจะตรวจร่างกายและตรวจเลือดดูความพร้อมของร่างกายว่าสามารถเข้าร่วมโครงการได้หรือไม่
2. แพทย์ผู้ทำการวิจัยจะเป็นผู้ชี้แจงให้ข้อมูลและตอบคำถามเกี่ยวกับโครงการวิจัยให้ท่านทราบก่อนที่จะเข้าร่วมโครงการ
3. ทารกที่เข้าโครงการจะถูกแบ่งออกเป็น 2 กลุ่ม คือ กลุ่มที่ได้รับยาน้ำ Ibuprofen ชนิดรับประทานและยาน้ำหลอก โดยได้ยารับประทานวันละ 1 ครั้งติดต่อกัน 3 วัน
4. แพทย์และเจ้าหน้าที่จะติดตามดูแลอาการเปลี่ยนแปลงบุตรของท่านและผลข้างเคียงที่อาจเกิดขึ้นอย่างใกล้ชิด
5. จะมีการเจาะเลือดเพื่อหาระดับยาในเลือดก่อนให้ยาและหลังให้ยา โดยมีการเจาะเลือดทั้งหมด 12 ครั้ง โดยในวันแรกเจาะทั้งหมด 8 ครั้งรวม 3.6 ซีซี ส่วนวันที่สองและวันที่สามเจาะวันละ 2 ครั้ง รวมวันละ1.4 ซีซีโดยการใช้ syringe ดูดจากสายสวนหลอดเลือดทางสะดือที่ใส่ไว้ในผู้ป่วยเพื่อผู้ป่วยจะไม่เจ็บตัวหลายครั้งจากการเจาะเลือดแบบทั่วๆไป
6. ผู้ป่วยทุกคนจะได้รับการตรวจ echocardiogram ก่อนและหลังได้รับยาแครบแล้วเป็นระยะ เพื่อยืนยันว่าไม่มีอาการของ PDA การทำ echocardiogram ดังกล่าวไม่มีความเสี่ยงแต่อย่างใด

ผลเสียที่อาจเกิดขึ้น

ทารกที่รับประทานยา Ibuprofen อาจมีผลข้างเคียงบางอย่างได้ เช่น ปัสสาวะลดลง ท้องอืด แรงดันหลอดเลือดในปอดสูงขึ้นได้ แต่การศึกษาในประเทศไทยที่ให้ยา ibuprofen ชนิดรับประทานที่โรงพยาบาลรามาธิบดี พบว่ามีทารกเพียง 1 คนจาก 32 คนที่มีท้องอืดแต่ไม่พบเลือดออกในกระเพาะ ส่วนการทำงานและหน้าที่ของไตไม่ลดลง นอกจากนี้หน้าที่การทำงานของตับก็ไม่เปลี่ยนแปลง ส่วนการศึกษาในต่างประเทศพบว่ายาไม่ได้มีผลทำให้ลดหน้าที่การทำงานของไต ไม่ได้ลดปริมาณเลือดที่ไปเลี้ยงลำไส้และไม่ได้ทำให้เลือดไปเลี้ยงสมองลดลงด้วย

ระหว่างการศึกษา ผู้ป่วยจะได้รับการดูแลในหอผู้ป่วยทารกแรกเกิด โดยมีแพทย์และพยาบาลติดตามดูอาการอย่างใกล้ชิด เช่น มีการตรวจติดตามสัญญาณชีพเป็นระยะๆ หากผู้ป่วยมีอาการเปลี่ยนแปลงที่ผิดปกติหรือเกิดภาวะฉุกเฉินสามารถรายงาน แพทย์หญิง วราภรณ์ แสงทวีสิน โทรศัพท์ 0-6060-5378 ได้ตลอด 24 ชั่วโมง

หากเกิดผลข้างเคียงหรือปัญหาแทรกซ้อนรุนแรงจากยาหรือเกิดอาการไม่พึงประสงค์จากการใช้ยาที่ไม่เคยมีรายงานมาก่อน จะหยุดทำการศึกษาในผู้ป่วยคนดังกล่าวทันทีพร้อมกับให้การรักษาที่เหมาะสมต่อไป

ผลประโยชน์ที่ผู้ป่วยจะได้รับ

1. เพื่อป้องกันการเกิดอาการ PDA เป็นการนำไปสู่การป้องกันและลดความเสี่ยงต่อการเกิดภาวะหัวใจล้มเหลว การหายใจล้มเหลว โรคลำไส้เน่าเปื่อย ไตวาย และลดความเสี่ยงต่อการเกิดพิษจากการใช้ oxygen
2. หลังจำหน่ายกลับบ้าน ผู้ป่วยจะได้รับการนัดหมายติดตามดูแลการเจริญเติบโตและพัฒนาการ รวมทั้งส่งเสริมป้องกันโรคอย่างต่อเนื่องเป็นระยะเวลาอย่างน้อย 1 ปี

การเข้าร่วมโครงการ

การเข้าร่วมโครงการวิจัยนี้ถือเป็นความสมัครใจ ไม่มีการบังคับใด ๆ ทั้งสิ้น การปฏิเสธที่จะเข้าร่วมโครงการไม่ถือว่าเป็นความผิดหรือสูญเสียประโยชน์ใด ๆในการรักษาทางการแพทย์ หลังจากที่บุตรของท่านจบโครงการวิจัยหรือออกจากโครงการวิจัยก่อนกำหนด ท่านจะยังคงได้รับการดูแลรักษาอย่างต่อเนื่อง

ค่าใช้จ่ายในการเข้าร่วมโครงการ

ในการให้ยา Ibuprofen การส่งตรวจพิเศษที่เกี่ยวข้องกับการใช้ยาและการตรวจเลือดเพื่อวัดระดับยา โครงการจะเป็นผู้ออกค่าใช้จ่ายทั้งหมด

ค่าชดเชยที่เกิดขึ้นจาการเข้าร่วมโครงการ

สถาบันสุขภาพเด็กแห่งชาติมหาราชินีจะรับผิดชอบในการรักษาทางการแพทย์จากอาการไม่พึงประสงค์ของยา Ibuprofen ที่ใช้ในการศึกษานี้ หรือจากขั้นตอนของโครงการวิจัยตามที่ทางสถาบันสุขภาพเด็กแห่งชาติมหาราชินี และผู้วิจัยกำหนดไว้ หรือการปฏิบัติที่ไม่ถูกต้องของเจ้าหน้าที่โรงพยาบาล แต่จะไม่รับผิดชอบต่อความผิดปกติที่สืบเนื่องมาจากโรคที่เป็นอยู่แล้วหรือการรักษาโรคนั้นๆ

การปกปิดความลับและการให้ข้อมูลจากการบันทึกทางการแพทย์

หากมิได้มีคำสั่งจากทางกฎหมาย จะมีได้เฉพาะแพทย์ผู้วิจัย คณะทำงานวิจัย และบุคคลที่ได้รับอนุญาตหน่วยงานทางราชการ รวมถึงคณะกรรมการจริยธรรมของโรงพยาบาลมีสิธิที่จะขอดูข้อมูลที่มีชื่อของท่านและบุตรได้ เนื่องจากจำเป็นที่จะต้องมีการเปิดเผยข้อมูลกับบุคคลที่กล่าวมาแล้วข้างต้น การปกปิดความลับดังกล่าวจึงอาจทำไม่ได้เต็มที่

ผลการศึกษาและข้อมูลอื่น ๆจากการวิจัยนี้ อาจนำเสนอต่อหน่วยงานราชการของประเทศไทย อย่างไรก็ตามชื่อของท่านและบุตรจะไม่ถูกเปิดเผยในรายงานหรือตีพิมพ์ต่อสาธารณชน เนื่องจากในการวิจัยนี้จะใช้เฉพาะชื่อย่อและหมายเลขประจำตัวผู้ป่วยแทนบุตรของท่าน

สิทธิของท่าน

ถ้าท่านไม่ต้องการให้บุตรของท่านเข้าร่วมโครงการศึกษาวิจัยนี้ บุตรของท่านยังคงได้รับการดูแลรักษาจากทางโรงพยาบาลตามปกติโดยไม่เปลี่ยนแปลง ท่านมีสิทธิ์ที่จะขอยกเลิกการเข้าร่วมโครงการศึกษาวิจัยครั้งนี้เมื่อไรก็ได้

ถ้าท่านต้องการให้บุตรของท่านเข้าร่วมโครงการศึกษาวิจัยนี้ ในการตัดสินใจเข้าร่วมโครงการ ท่านควรทราบเงื่อนไข ดังต่อไปนี้

ก) การตัดสินใจเข้าร่วมโครงการ มีท่านเป็นผู้ตัดสินใจเอง

ข) ถ้าท่านตัดสินใจเข้าร่วมโครงการ แต่ว่าต่อมาเปลี่ยนใจ ท่านสามารถยกเลิกการเข้าโครงการเมื่อไรก็ได้

ค) ถ้าท่านตัดสินใจไม่เข้าร่วมโครงการ บุตรของท่านจะยังคงได้รับการดูแลตามมาตรฐานการรักษาจากโรงพยาบาลแห่งนี้ตามปกติ

บทสรุป

ก่อนที่ท่านจะให้คำยินยอมโดยการลงลายมือชื่อในเอกสารฉบับนี้ ท่านมีโอกาสที่จะสอบถามเกี่ยวกับขั้นตอนการทำวิจัย และยาที่ทำการวิจัย ผลข้างเคียงจากการใช้ยา จากข้อมูลทั้งหมดที่ท่านได้รับนี้ ท่านสมัครใจที่จะให้บุตรของท่านเข้าร่วมโครงการศึกษาครั้งนี้ ท่านยังสามารถสอบถาม หากมีข้อสงสัยใด ๆ ในระหว่างที่เข้าร่วมโครงการวิจัย ท่านต้องแน่ใจว่าท่านได้อ่านเอกสารคำยินยอมฉบับนี้แล้ว การลงลายมือชื่อในเอกสารยินยอมนี้แล้ว หมายถึงท่านยินยอมที่จะให้นำข้อมูลที่เกี่ยวข้องกับการเข้าร่วมวิจัยในบันทึกข้อมูลทางการแพทย์ของบุตรของท่านให้แก่ คณะผู้วิจัยจะนำข้อมูลเพื่อใช้ในการตีพิมพ์ทางด้านการแพทย์ หน่วยงานทางราชการ อย่างไรก็ตามชื่อของบุตรของท่านจะไม่มีการเปิดเผยในรายงานผลการศึกษา

โครงการศึกษาวิจัย

เรื่อง

การใช้ยา Ibuprofen ชนิดรับประทาน ในการป้องกันการเกิด symptomatic patent ductus arteriosus ในทารกคลอดก่อนกำหนด(Oral ibuprofen suspension administration to prevent symptomatic patent ductus arteriosus in premature infant)

เอกสารแนะนำสำหรับอาสาสมัคร

- ข้าพเจ้าได้รับการชี้แจงวัตถุประสงค์ของโครงการ ขั้นตอนการปฏิบัติหลังการเข้าร่วมโครงการ ผลดีและผลเสียของการเข้าร่วมโครงการอย่างละเอียดครบถ้วนแล้ว ข้าพเจ้าสามารถถามคำถามใดๆก็ได้เกี่ยวกับการเข้าร่วมโครงการและทุกๆคำถามของข้าพเจ้าได้รับคำตอบแล้ว
- ข้าพเจ้ายินยอมให้บุตรของข้าพเจ้าเข้าร่วมโครงการนี้ ข้าพเจ้าเข้าใจด้วยว่าข้าพเจ้าอาจขอให้บุตรของข้าพเจ้ายกเลิกการเข้าร่วมโครงการเมื่อใดและด้วยเหตุผลใดๆก็ได้ ข้าพเจ้าเข้าใจด้วยว่าถ้าข้าพเจ้ายกเลิกการเข้าร่วมโครงการ จะไม่เกิดผลเสียใดๆอันเนื่องมาจากการยกเลิกเข้าร่วมโครงการและบุตรของข้าพเจ้าจะยังคงได้รับบริการทางการรักษาตามปกติจากโรงพยาบาลนี้

ลงนาม ..... ผู้ยินยอมให้เข้าร่วมโครงการวิจัย

(.....)

.....

ลายพิมพ์นิ้วหัวแม่มือ*

เกี่ยวข้องเป็น.....ของผู้ป่วย

วันที่ ...../...../.....

* ในกรณีที่ผู้ให้ความยินยอมไม่สามารถอ่านหนังสือแสดงความยินยอมนี้ได้ ขอยืนยันว่าหนังสือฉบับนี้ได้ถูกอ่านและอธิบายข้อความให้ผู้เข้าร่วมโครงการับฟังอย่างถูกต้องโดยผู้วิจัยและผู้เข้าร่วมโครงการได้พิมพ์นิ้วหัวแม่มือแสดงความยินยอม

ลงนาม.....พยาน

(.....)

วันที่ ...../...../.....

ลงนาม.....พยาน

(.....)

วันที่ ...../...../.....

ตามที่ผู้ปกครอง/ผู้ดูแลผู้ป่วยตามกฎหมายได้ลงลายมือชื่อลงในเอกสารฉบับนี้แล้ว ข้าพเจ้าแพทย์ผู้ดูแลขอยืนยันว่า ได้อธิบายให้ผู้ปกครอง/ผู้ดูแลตามกฎหมาย ของผู้ป่วยทราบโดยละเอียดถึงลักษณะโครงการ ความเสี่ยงและประโยชน์ที่จะได้รับจากการที่ผู้ป่วยเข้าร่วมในโครงการศึกษา ข้าพเจ้าได้มอบสำเนาหนังสือฉบับครบสมบูรณ์นี้รวมทั้งหมายเลขโทรศัพท์สำหรับติดต่อให้ผู้เข้าร่วมโครงการเรียบร้อยแล้ว

ลงนาม ..... แพทย์ผู้ทำการวิจัย

(.....)

วันที่ ...../...../.....

หากท่านมีข้อสงสัยประการใดอันอาจเกิดขึ้นในขณะนี้หรืออาจเกิดขึ้นภายหลัง กรุณาสอบถามได้จากคณะผู้ทำการวิจัยโดยตรง ณ สถานที่และหมายเลขโทรศัพท์ดังต่อไปนี้

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