Paediatric and Perinatal Drug Therapy **Volume 4, Issue 4, 2001**

European Society for Developmental Pharmacology, 7th Congress

The 7th Congress of the European Society for Developmental Pharmacology met on November 17th–20th 2001 in Limassol, Cyprus. The meeting was a great success despite having had to be postponed from Jerusalem the previous year. The President for the meeting was Rafael Gorodischer. There was an interesting programme covering different aspects of drug evaluation in children, pain and analgesia, prostaglandins and COX inhibitors, genetics and pharmacotherapy, anti-infective therapy and vaccines, ethics, licensing of medicines and the need for a register of clinical trials in children. A global perspective was given in relation to pharmaco-economics and paediatric pharmacotherapy in the developing world.

There were 14 oral free communications and 21 poster presentations. These are shown below (O = oral, P = poster).

01
Maternal Smoking during Pregnancy: Quantification of Fetal Exposure by Hair Analysis and Newborn Effects

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Smoking during pregnancy is a major health problem. In this study, our aims were to quantify maternal smoking and fetal exposure by hair analysis for nicotine cotinine and to identify risk factors for fetal and neonatal adverses effects. 254 pregnant women, reporting to smoke at least one cigarette per day were included during the first trimester of pregnancy and followed prospectively until delivery. A structural questionnaire on general health, obstetrical history and clinical features was completed and a detailed history of smoking habits during pregnancy was recorded. At birth, hair samples were obtained from the mother and the baby. Nicotine and cotinine concentrations were measured by radioimmunoassay (Brandeis University, Mass. USA). 187 mother-infant pairs were analyzed. Mothers, aged 22 to 42 years, were classified in 5 groups according to the number of cigarettes per day during the third trimester: no smoking (4.8%), 1–5 (24.6%), 6–10 (29.9%), 11–15 (20.9%), 16 and over (19.8%). Concentrations of nicotine and cotinine were 30.8 ± 23.1 and 2.10 ± 2.34 ng/mg hair, significantly related to smoking ($P<0.0003$ and $P<0.001$). The infants weighted 1600 to 4650 g and had a gestationel age of 34 to 42 weeks. Nicotine and cotinine concentrations were 5.51 ± 6.20 ng/mg and 1.20 ± 1.06 ng/mg hair. Using a multivariate analysis, the hair concentrations of cotinine in the baby was significantly related to maternal tabacco consumption ($P<0.01$), maternal hair concentrations of cotinine ($P<0.001$) and risk of premature delivery ($P<0.04$). Hair cotinine (but not nicotine) concentrations in the infants born from smoking mothers were significantly related to maternal smoking and risk of premature delivery. The correlation with other perinatal risks is currently under investigated in a larger population.

02
Evidence of Cisapride Metabolism Immaturity in Neonates

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Cardiac toxicity of cisapride has been reported in neonates without . any of the risk factors reported in children or adults. Therefore, age per se may be a risk factor. Our hypothesis was that CYP3A7 (the major CYP isoform expressed in neonatal liver) has no or little activity toward cisapride resulting in the inability of the neonatal liver to eliminate cisapride. Incubations were performed with Ad 293 cells transfected with full length CYP3A4, CYP3A5, CYP3A 7, CYPIAI, CYPIA2, CYP2C8, CYP2C9 cDNA inserted into expression vector and with human hepatic microsomes from fetuses, neonates, infants and adults. Microsomal preparations were characterised for total cytochrome P450 and their individual content in CYPIAI, CYPIA2, CYP2C, CYP2D6, CYP2EI, CYP3A, and CYP4A were determined by immunoblotting after SDS-gel electrophoresis. After incubation, the culture medium was removed and processed for analysis by HPLC. The biotransformation of cisapride was mediated by CYP3A4 (the major CYP expressed in adult liver) but not by CYPIAI, CYPIA2, CYP2C8, CYP2C9, CYP3A5 and with a minor contribution of CYP3A 7. Using microsomes, norcisapride formation was significantly correlated with CYP3A4 activity as measured by testosterone 6 hydroxylase (a specific marker of CYP3A4) but not with hydroxylation of dehydroepiandrosterone, a reliable index of CYP3A 7 activity. Microsomes of foetuses and neonates less than I week deprived of CYP3A4 had no or negligible activity toward cisapride. Thereafter the metabolism of cisapride steadily increased to reach activities exceeding adult values. Our results demonstrate that the metabolism of cisapride is decreased during the first week of life and help to relate the toxicity of this drug in neonates to an immaturity in cisapride metabolism.

03
Effect of Adenosine AI Receptor Blockade on the Hypoxemia-Induced Renal Vasoconstriction of the Newborn Rabbit

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Background: Hypoxemic conditions encountered in newborn infants can lead to renal insufficiency. We previously demonstrated the major role of adenosine in the hypoxemia-induced acute renal failure in newborn rabbits. The purpose of the present study was to further define the role of adenosine in this model by using the specific adenosine AI-receptor antagonist 8- cyclopentyl-I,3-dipropylxanthine (DPCPX).

Methods: Experiments were performed in 24 anaesthetized and mechanically ventilated newborn rabbits. Renal function and hemodynamic parameters were measured in normoxemic (Nx)(PaO_2 ~ 100 mmHg) and hypoxemic (Hx) conditions (PaO_2 ~ 40 mmHg) during a control period and two treatment periods during which DPCPX was infused at 60 ~g/kg/h.

Results: In Nx, DPCPX administration induced a significant increase in urine flow rate (UV) (+36%) with an increase in glomerular filtration rate (GFR)(+15%) despite a significant decrease in renal blood flow (RBF((-20%) and a significant increase in renal vascular resistance (RVR)(+30%). In Hx, RBF was significantly decreased (-35%) with an increase in RVR (+40%); GFR and UV were not significantly influenced by DPCPX. In both groups, a significant increase in filtration fraction (FF) occurred and sodium excretion was increased by DPCPX (+ 105% in Nx and + 132% in Hx).

Conclusion: The overall results demonstrate an increase in the renal vascular resistance in the immature renal vascular bed after A 1 blockade with DPCPX, the increase in FF suggesting preferential efferent vasoconstriction. This supports an Al receptor-mediated efferent vasodilatory effect of adenosine in physiologic conditions. DPCPX failed to prevent the Hx- induced renal hemodynamic changes: the contribution of adenosine to the adverse effects of hypoxemia is not mediated via A 1 receptors.

04
Midazolam as CYP3A Probe in Preterm Infants

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Background: CYP3A4 activity is low directly after birth increasing rapidly in the first two weeks of life. Midazolam (M) is mainly metabolized by CYP3A4 to 1-OH-M. Therefore M is used as CYP3A4 probe in adults. We studied the PK of intravenous M and 1-OH-M in preterm infants and studied the possibility of M as CYP3A probe in these children.

Methods: Midazolam (0.1mg/kg) was administered to 25 preterm infants (GA 25-33 wks) as a 30-minute IV infusion. Blood samples (0.2ml) were taken for M and 1-OH-M concentration measurements at 0, 0.5, 1, 2, 4, 6, 12 and 24 h after M administration. M and 1-OH-M concentrations were measured using GC-ECD with a lower limit of detection of 1ng/ml (M) and 0.2ng/ml (1-OH-M). Serum-concentration time curves of M were analyzed using a weighted least-squares simplex algorithm (weight 1/Y2). Relationships between PK parameter estimates, M concentrations and demographic parameters were determined with Spearman's correlation test.

Results: M concentration-time data best fitted a one-compartment model. Cl was 0.14±0.09 l/kg/h, t1/2 6.8±3.7 h and Vss 1.24±0.8 l/kg. 1-OH-M concentrations could be detected in 14 out 25 patients, C_{max} 18±28 ng/ml, Tmax 6.3±6.3h. No relationship was found between bodyweight corrected CL and postnatal, postconceptional or gestational age. However, volume of distribution and for body weight corrected clearance correlated significantly with postnatal indomethacin exposure.

Conclusion: Consequent to immature hepatic CYP3A4 activity,midazolam clearance and 1-OH-midazolam concentrations are markedly reduced in preterm infants as compared to previous reports from studies in older children and adults. Indomethacin exposure and its apparent impact on midazolam clearance supports alteration of drug disposition produced by a patent ductus arteriosus or by changes in hemodynamics or renal function. This study illustrates the importance of other co-variates on midazolam clearance. Therefore, midazolam clearance as CYP3A probe in preterm infants should be used with caution.

Supported by grant 216, Sophia Foundation for Research Rotterdam, Erasmus University Trust Rotterdam and Roche, Inc, USA

05
PK of Oral Midazolam in Preterm Infants

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Background: Intravenous Midazolam (M) is frequently used as a sedative in preterm infants at Neonatal Intensive Care Units. In older children at the intensive care, M is also administered orally, especially in cases where intravenous access is difficult or impossible. However, no pharmacokinetic data are available of oral midazolam administration in preterm infants, by which dosing schedules might be calculated. *Methods*: Midazolam (0.1mg/kg, in 0.5ml glucose 5%) was administered to 15 preterm infants (GA 25-33 wks) via the gastric tube. Blood samples (0.2ml) were taken for M and 1-OH-Midazolam (1-OH-M) concentration measurements at 0, 0.5, 1, 2, 4, 6, 12 and 24 h after M administration. M and 1-OH-M concentrations were measured using GC-ECD with a lower limit of detection of 1ng/ml (M) and 0.2ng/ml (1-OH-M). Serum-concentration time curves of M were analyzed using a weighted least-squares simplex algorithm (weight 1/Y2). Results M concentration-time data best fitted a one-compartment model. Cl/F was 0.24±0.0.2l/kg/h, $t_{1/2}$ 5.1±3.1 h and Vss 2.8±5.0 l/kg. Mean M plasma peak concentration was 63.5±45.9 ng/ml and t_{max} 2.9±2.5h. 1-OH-M concentrations above the detection limit were only measured in 9 out of 15 patients with C_{max} 7.0±8.0 ng/ml and t_{max} 13±10h.

Conclusion: This is the first report on M disposition after oral administration to preterm infants. Oral M clearance rate, normalized for body weight, is only 10% when compared to children older than 6 months of age[1]. Moreover, in comparison to older childern mean M t_{max} was longer with higher mean peak serum concentrations in preterm infants. These age-related differences in oral midazolam disposition are likely explained by reduced hepatic and intestinal CYP3A4 activity in newborns.

Reference

1. Payne K *et al*. Eur J Clin Pharmacol 1989;37:267-272.

Supported by grant 216, Sophia Foundation for Research Rotterdam and Roche, Inc, USA

06
Enantiospecific PK/PD-Modeling of Heart Rate and Mean Arterial Pressure During Carvedilol Treatment in Children with Congestive Heart Failure

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Background: Reduction of heart rate (HR) and mean arterial pressure (MAP) under beta-receptor blocker therapy contribute to the improvement in ventricular function of children with heart failure. Beta-receptor blockade is predominantly caused by the S-enantiomer of carvedilol, whereas both enantiomers are equipotent alpha-blockers. For optimizing therapy in pediatric patients, investigations of potential developmental differences in beta-blocker response are fundamental.

Methods: Sixteen pediatric patients with heart failure received carvedilol (C) (0.09, 0.18, 0.36 for 14 days each, target dose 0.7 mg/kg per day) in addition to digoxin, ACE-inhibitor and diuretics. HR, MAP, racemic and enantiomeric C plasma concentrations were monitored after the initial and the maximum dose level during one dosing interval. Potency (EC_{50}) and maximum reduction (E_{max}) of HR and MAP towards C were determined by pharmacokinetic/ pharmacodynamic modeling (PK/PD-modeling).

Results: The infants and toddlers (IT, 6 weeks to 3.5 years, *n*=8) were more sensitive to HR reduction after C than the children and adolescents (CA, 5.5 to 19 years, *n*=8). In contrast, C was more effective to reduce blood pressure in the infants and toddlers than in the older pediatric patients.

Conclusion: Beta-receptor blocker-induced HR- and MAP-reduction are differently regulated during childhood development. Differences in potency and efficacy of carvedilol result in an earlier onset of protection from excessive adrenergic stimulation in the young compared to the older pediatric patients.

This study is supported by the German Heart Foundation.

Patients	Heart rate		Mean arterial pressure	
	EC_{50} of C (ng/ml)	E_{max} (% reduction)	EC_{50} of C (ng/ml)	E_{max} (% reduction)
IT	3.6 ± 2.0*	21.1 ± 8.2	5.6 ± 3.5	33.3 ± 12.1*
CA	10.8 ± 1.8	20.6 ± 9.0	4.0 ± 2.6	20.5 ± 11.1

Values are means ± SD, **P*<0.05 versus children and adolescents. The age-dependent potency of C for heart rate reduction was confirmed by PK/PD-modeling with the S-carvedilol concentrations.

07
Renal Effects of Cyclooxygenase- Type-2 (COX-2) Inhibition with Nimesulide in the Newborn Rabbit

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Background: Tocolysis with non-steroidal anti-inflammatory drugs (NSAIDs) has been widely accepted for several years. Recently, the use of cyclooxygenase 2 (COX-2) selective NSAIDs such as nimesulide has been proposed. However, recent data reporting neonatal acute renal failure (ARF) or irreversible end-stage renal failure after maternal ingestion of nimesulide question the safety of this drug in pregnancy. Therefore, this study was designed to define the renal effects of the COX-2 selective NSAID nimesulide in newborn rabbits.

Methods: Experiments were performed in 20 anaesthetized and mechanically-ventilated newborn rabbits. Renal function and hemodynamic parameters were measured using inulin and P AH clearances as markers of glomerular filtration rate (GFR) and renal blood flow (RBF), respectively. After a control period, nimesulide, 200 or 400 µg/kg, was given as an iv bolus, followed by a 5 µg/kg/min infusion throughout the experiment.

Results: Nimesulide 200 and 400 µg/kg administration induced a significant increase in renal vascular resistance (RVR) (+112 and +86%; *P*<0.01 and 0.05 respectively), with a concomittant decrease in GFR (-48 and -40%; both *P*<0.01), RBF (-53 and -42%; *P*<0.01 and 0.05) and diuresis (-41 and -42; both *P*<0.01). Mean arterial blood pressure (MAP) and filtration fraction (FF) remained unchanged after nimesulide administration.

Conclusion: The overall results demonstrate an increase in the RVR in the immature renal vascular bed after COX-2 inhibition with nimesulide. The stable FF indicates both pre- and postglomerular vasoconstriction, leading to oliguric ARP. These *Results*: confirm that prostaglandins, by maintaining an afferent and efferent vasodilatory tone, play a key role in the preservation of glomerular filtration in the neonatal period. They do not support the claim that COX-2 selective inhibition may be safer for the kidney than non selective NSAIDs

08
Indomethacin Treatment of Patent Ductus Arteriosus (PDA) in Preterm Infants: Eight Years Experience with the Known, Standard Indomethacin Treatment Regimen

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Background:. Our experience with indomethacin treatment of PDA is not in line with recently published studies demonstrating a failure in 30% or more of indomethacin treated infants together with renal side effects.

Methods:. Retrospective analysis of the hospital charts of all preterm infants with a gestational age ≤32 weeks and a diagnosis of PDA who were treated in our hospital from June 1992 to May 2000. The hospital charts were analyzed with respect to the hemodynamic significance of the PDA and the efficacy of the treatment. Treatment was considered successful I, if echocardiography demonstrated PDA closure or a PDA with little hemodynamic significance together with clinical improvement of the patient (i.e. improved ventilator settings). In a subset of indomethacin treated infants born after June 1995, the charts were analyzed for the indomethacin dosage regimen used, and renal and gastrointestinal side effects.

Results: 412 preterm infants were identified. A PDA was present in 76 infants (18 %; gestational age median 28, range 23–32 weeks; mean birth- weight 1044 (SD 369) g. In 20 infants with a PDA of little hemodynamic significance, the PDA closed spontaneously. The remaining 56 infants (gestational age median 27, range 23–31 weeks; birthweight mean 936, SD 262 g) were treated with indomethacin. Indomethacin was effective in 51 infants (91 %). In one infant, the PDA was surgically ligated after reopening. Four infants died from complications of prematurity. Preceding death, PDA was hemodynamically significant in two infants, and almost closed and of little hemodynamic significance in the remaining two infants. The majority of infants (57 %) was treated with the standard indomethacin dosage regimen (three doses of 0.2 mg/kg every 12 hours), 11 % of infants received less than three doses, and 32% of infants were treated with four doses or more. Besides a transitory rise in serum creatinine and oliguria during indomethacin treatment, no major renal or gastrointestinal side effects were observed.

Conclusion: Indomethacin is a highly effective and safe treatment for PDA in preterm infants. If indomethacin is appropriately used, the need for surgical ligation or the introduction of alternative drugs is low.

09
Maturation of Zidovudine Glucuronidation in Neonates and Infants

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Zidovudine is commonly administered during pregnancy and the neonatal period to prevent mother-to-child HIV -1 transmission. While clearance of zidovudine was demonstrated to be reduced in neonates, no description of maturation of the relevant metabolic pathway was available. We used the data set from 100 children and 27 adults involved in AZT therapeutic monitoring to estimate the maturation of ZDV glucuronidation in human. Neonates and infants received 2 to 4 mg/kg orally or intravenously. Adult doses were 300 mg. The ZDV -glucuronide/ZDV concentration ratio was determined by HPLC. No influence of gender or gestational age was detected. G-ZDV/ZDV ratio was very low at birth (approximately 2) and dramatically increased to reach adult value (approximately 9) after one month of age. Consistently, plasma concentrations of ZDV were much higher in neonates than in infants and adults. When the achieved plasma concentrations were normalized to the target concentration used for therapeutic drug monitoring (i.e. mean plasma concentration obtained in a reference adult group) a significant relationship was evidenced between the metabolic G- ZDV/ZDV and the measured/target concentration ratio (r=0.25, *P*=0.01). These results suggest 1) that the maturation of ZDV glucuronidation occurs during the first month of life 2) immature glucuronidation is related to lower ZDV elimination 3) lower ZDV doses normalized to body weight should be given to neonates to achieve plasma concentration similar to older children and adults consistently with previous population pharmacokinetic data (Mirochnich *et al*., CPT, 1999)

010
Neflinavir doses should be increased in infants less than 3 months

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Perinatally HIV-infected infants with high viral load in the first months of life are at risk for a rapid progression of disease and early antiretroviral treatment could prevent such deterioration. Nelfinavir in combination with didanosine and stavudine in infants less than 3 months is currently evaluated as part of a phase I/II trial conducted by the Pediatric European Network for Treatment of AIDS (PENTA). In this study, the dose of nelfinavir (Viracept*) was initially 40 mg/kg thrice daily (TID) as powder or tablet (120 mg/kg/d) but was increased to 75 mg/kg twice daily (BID) (150 mg/kg/d). Didanosine and stavudine were both administered twice daily at the dose of 100 mg/m^2 and 1 mg/kg respectively. The pharmacokinetic study (6 blood samples) was performed in 8 patients, aged 1.5 to 7.2 months at least 2 weeks after initiation of the triple therapy. Plasma concentrations of nelfinavir were measured by HPLC-MS. The daily dose of nelfinavir ranged between 117.2 and 150 mg/kg with equal number of patients receiving tablet or powder. The CL/F and the Vd/F were 4.0 l/kg/h ± 0.7 [2.5–4.5] and 18.2 l/kg ± 8.9 [8.8–22.3]. The $t_{1/2}$ was 3.5 hours [1.4–6.2] (mean and SD). Important interindividual variability was observed for C_{min} without correlation with the daily dose of nelfinavir or the patient's age. The minimal therapeutic C_{min} established in adults (1000 ng/ml) was reached in only 2 patients and only infants receiving 130–150 mg/kg/d nelfinavir achieved the adult target value for $AUC_{0\text{-}24\ h}$ (≥ 30000 ng/ml/h). Infants less than 3–4 months need higher doses (130–150 mg/kg/d) of nelfinavir compared to older children and adults to achieve therapeutic concentrations. Decreased nelfinavir bioavailability is a possible explanation for the results observed in this age group.

011
Population Pharmacokinetics of Cisapride in Preterm and Term Neonates

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Background: Cisapride is used in neonates with gastro-oesophageal reflux despite scant pharmacokinetic evaluation in this population. In addition, concentration-dependent toxicity has been described and metabolic immaturity in neonates may lead to increased concentrations in this population.

Methods: We conducted a population pharmacokinetic analysis of cisapride in neonates using NONMEM. Cisapride was administered orally at the dose of 0.2 mg/kg four times a day. Blood samples were drawn at the same time than another biological control. Plasma concentrations were measured using a validated HPLC method. Ninety-one subjects 26–37 weeks of gestational age and 750–2780g birth weight were included. Two-hundred fifty concentrations were obtained from 2 to 123 days of life. Times of sampling varied from 10 min to 13.5 h after dosing.

Results: Cisapride concentrations ranged from 5.5 to 172 ng/ml and were not higher than those reported in adults. The data were fitted using a one-compartmental model with first-order absorption. Since too few samples were drawn early after dosing, the absorption constant was fixed to 2.5/h. Clearance (CL/F) was significantly related with weight. No significant relationship was found between volume of distribution (V/F) and covariates. Final population pharmacokinetic parameters (interindividual variability) were: V/F=18.4L (25.2%) and CL/F=0.504*WT + 204 ml/h (38.1%).

Conclusion: Our finding that cisapride clearance is primarily influenced by weight is in agreement with current recommendations of weight-adjusted doses. However, target concentrations are not known in neonates and could be different from those in adults.

012
Pharmacokinetics and Efficacy of Lanzoprazole in Children with Peptic Oesophagitis

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Lansoprazole is a proton pump inhibitor extensively used in adults (daily dose: 30 mg). The present study was designed to define the pharmacokinetics and dose required in children. Twenty-three children (11 girls) aged 4 months to 13 years and requiring lanzoprazole for oesophagitis were studied, after parental written informed consent. Lanzoprazole (17 mg/m^2) was given once daily and after 7 days of treatment, a 24-hour gastric pH and a pharmacokinetic study were performed. The daily dose was considered effective if the gastric pH was >3 during more than 65% of the 24 hour period. If it was less than 65%, the dose of lansoprazole was doubled and at day 14, a second 24-hour intragastric pH was performed. If lanzoprazole was judged ineffective, the treatment was modified, while, in the patients who responded, the treatment was maintained. Under 17mg/m^2 (0.73mg/kg, range 0.54 – 0.91), 9 (39%) children had a gastric pH >3 during more than 65% of the 24-hour gastric pH (group 1). 14 patients received the doubled-dose of 30.4 mg/m^2 (1.44mg/kg, range 1.18–1.66, group 2) and lanzoprazole was effective in 6 (26%). In the 23 patients, AUC was 1178 ± 1295 ng/h/ml and $t_{1/2}$ was 1.0 ± 0.4 h (mean, SD). However, AUC (ng/h/ml) was different between the 2 groups (2035 ± 559 vs 627 ± 138 ng/h/ml, P<0.01) but Cmax (641 ± 136 and 350 ± 75 ng/ml, P=0.05) and oral clearance (0.76 ± 0.25 vs 2.55 ± 0.73 l/h/kg, P=0.07) were similar (mean, SEM) Lansoprazole appears safe and effective in children. Sixty-five percent of children responded to 1.44mg/kg or less. The responders to the lower dose had higher AUC, suggesting that variability in drug response may be related to pharmacokinetics.

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013
Antipyretic Efficacy of an Initial 30 mg/kg Loading Dose of Acetaminophen versus a 15 mg/kg maintenance dose

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Objective: To compare the antipyretic efficacy an initial loading dose of acetaminophen 30 mg/kg vs 15 mg/kg.

Design: Double-blind, parallel group, randomized clinical trial Participants: 121 febrile (defined as rectal temperature of 39°C to 40 °C) but otherwise healthy outpatients, weighing from 4 to 26 kg were randomly assigned to one of the studied dose (62: 15 mg/kg and 59 : 30 mg/kg).

Results: The average time to obtain a temperature lower than 38°5 was significantly shorter in the group receiving 30 mg/kg vs 15 mg/kg (110 ±94 min vs 139 ±113 min) using "an intention to treat analysis". The maximum temperature decrease was significantly higher in the group receiving 30mg/kg compared to 15 mg/kg (2.3 ± 0.7 °C vs 1.7 ±0.6 °C). The mean time with temperature below 38°5 was significantly longer in the 30 mg/kg group compared to the 15 mg/kg group (250±92 min vs 185 ±121 min respectively). Adverse events were reported in 5 children in the 30 mg/kg group compared to 6 in the 15 mg/kg group/ (hyperthermia, hypothermia, vomiting). The difference was not statistically significant.

Conclusion: An initial 30 mg/kg loading dose of acetaminophen appeared to be more effective in reducing fever than a 15 mg/kg maintenance dose. No difference was observed regarding tolerance. These data suggest that acetaminophen treatment of fever may be more efficient being initiated using an initial loading dose.

014
Thiopurine s-Methyltransferase Modulates the Effects of 6-Mercaptopurine in Childhood Acute Lymphoblastic Leukemia

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The major metabolic pathway that catabolizes 6-mercaptopurine (6-MP) is methylation through thiopurine S-methyltransferase (TPMT), a cytosolic enzyme liable to genetic polymorphism. As far as an optimal maintenance therapy using 6-MP remains crucial in the prognosis of ALL, 78 children with ALL were studied during maintenance to investigate the relation between the pharmacogenetics of TPMT and the effects of 6-MP initially given at a daily dose of 50 mg/m^2 and adapted to maintain WBC in the cytotoxic target of 2000-3000 cells/mm^3. Mean 6-MP dosage ranged from 27 to 123 mg/m^2/d and mean TPMT activity from 3.3 to 41.2 nmol/h/ml. Patients with high TPMT activity received higher 6 MP doses compared to patients with a low TPMT activity, presented higher WBC, lower percentage of leucocyte counts within the cytotoxic target (P<0.0001) and higher occurrence of infection episodes (P<001). In a subgroup of 26 patients, the methylated thiopurine nucleotides of 6-MP (Me6-MPN) and 6-thioguanine nucleotides (6-TGN) concentrations in red blood cells were measured. The Me6-MPN/6-TGN ratio was positively correlated with the 6-MP dosage (P=0.004) and TPMT activity (P=0.01). Therefore, a predominance of the methylated metabolites of 6-MP over the non methylayed metabolites in patients having a high a high TPMT activity could explain the dissociation between the cytotoxic and immunosuppressive effects of 6-MP. Pharmacogenetics of TPMT should already be taken into account to optimize the maintenance therapy in ALL.

P1
Unlicensed and Off-label Prescribing of Drugs for Children in General Practice

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Background: Hospital based studies have shown that many drugs used in children are either not licensed (unlicensed) or are prescribed outside the terms of the product license (off-label). The majority of children are seen by doctors in general practice in the UK and we wish to determine the incidence and nature of unlicensed and off label prescribing of drugs for children in this setting.

Methods: A retrospective analysis of all prescriptions for one year involving children ≤ 12 years of age from a single suburban general practice in the English Midlands. Prescribed drugs were categorised as licensed, unlicensed (ie without a product licence) or used in an off label way (ie outside the terms of their product licence).

Results: During 1997 there were 3347 prescription items involving 1175 children ≤12 years old and 160 different drugs. 2828 (84.5%) prescriptions were for licensed medicines used in a licensed way; 10 (0.3%) were unlicensed; 351 (10.5%) were licensed medicines used in an off label way. For 158 (4.7%) the information was insufficient to determine licence status.

Conclusion: A significant number of drugs prescribed for children by general practitioners are off label and highlights the anomalies and inadequacies of drug information for prescribers.

P2
Use of "Off-Label" and Unlicensed Drugs in Neonatal and Paediatric Intensive Care Units in France

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In paediatric practice, drugs that are either unlicensed for use in children or are prescribed outside the terms of the product license ("off-label") are often prescribed. There is, however, little information available on the extent to which these types of treatment are used. We therefore studied unlicensed drug use prospectively. Prescriptions for all patients hospitalised in 59 French neonatal and paediatric intensive care units over a 24 hours period were reviewed. Each drug was assessed according to the French product licence. 2383 courses of drugs were administered to 588 patients (4.6/patient). 49.6 % of the patients were less than 1 month. 40.2 were premature. 65% of the drug courses (prescriptions) were unlicensed for use in children and 13%, 28%, 9% were off-label in relation respectively to the dose, the indication and the route of administration. 73% were unlicensed or off-label for more than 1 reason. The total of unlicensed for age and off-label prescriptions were 94%. The 10 most frequently administered unlicensed or off-label drugs were caffeine citrate, midazolam, fentanyl, cefotaxime, furosemide, propacetamol, amoxicilline, amikacin, vancomycin, domperidone. The percentage of unlicensed and off-label drug prescriptions decreased with increasing age. There was no specific paediatric dosage for 73% of the prescriptions. In conclusion, many of the drugs extensively prescribed in French neonatal and paediatric intensive care units were not approved by the licensing authority.

P3
Unapproved Prescriptions in Two Paediatric Intensive Care Units in Israel

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Background. Solutions to the "therapeutic orphan" issue are currently implemented in the United States, and Europe is searching for its frame of action to its contribution. Those efforts will effect pediatric therapeutics worldwide. Main considerations in setting priorities for studying medications in children are their therapeutic value and extent of use. In this study we evaluated the extent of unlicensed and off-label use of medications in Pediatric Intensive Care Units (PICU) of two hospitals: Soroka (SH) and Assaf Harofe (AHH), in Israel.

Methods: Medical records of 158 patients treated in PICU of 2 hospitals in Israel in representative seasons of the year 2000 were reviewed. Commonest diagnoses included head trauma, infectious illnesses, seizures and asthma.

Results: The total number of different medications used was 124. Sedatives and antibiotics were the most frequently prescribed medications in SH, and antibiotics, paracetamol and antiasthmatics in AHH. Sympathomimetics, sedatives, antiasthmatics and antibiotics were the most common off-label/unlicensed medications categories used. Eighty-three percent of patients received unlicensed/off-label medications, and 42% of prescriptions were unlicensed or off- label. Inappropriate age was the most common off- label category, followed by different indication, different route, and different dose. Differences were observed between the two PICU; they may relate to different therapeutic practices and/or to different pathologies.

Conclusion: Out of necessity, a majority of the sickest children in Israel are exposed today to medications that have not been appropriately studied in the pediatric population. Our *Results*: are consistent with a British study and indicate an urgent need to investigate those medications in children.

P4
Population Pharmacokinetic Analysis of Netilmicin in Neonates and Infants Using a Nonparametric Method

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Background: Although the therapeutic and toxic effects of netilmicin are related to its plasma concentration, its pharmacokinetics in neonates and infants and the influence of clinical and biological variables have been only partially assessed.

Methods: Therapeutic drug monitoring data collected from 186 neonates and 95 infants receiving netilmicin were analyzed with a nonparametric (NPML) population approach. The influence of gestational and postnatal age, weight, Apgar score, and creatinine and urea plasma concentrations on the pharmacokinetic parameters was assessed. The neonate and infant groups were each randomly divided into a learning sample and a validation sample. The population analysis was performed on each learning subgroup with the NPML method. In the validation group, the data were used to assess the concentration predictability. Since there is no specific netilmicin formulation for neonates and infants, an error model was proposed to account for errors due to dilution processes when preparing the infusion.

Results: In neonates, the covariates that reduced expected variance of plasma clearance by more than 10% were postnatal age, body weight and plasma creatinine, and in infants, plasma urea and creatinine. Body weight and sex played a significant role in explaining the variability of the volume of distribution. The accuracy of the concentration predictability assessed in the validation samples were satisfactory, and no significant bias was found.

Conclusion: These findings help explain the large interindividual variability of the pharmacokinetics of netilmicin and the influence of the clinical and laboratory covariates in neonates and infants.

P5
Renal Toxicity with Aciclovir in an Adolescent

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We report a case of renal toxicity during treatment with acyclovir, probably due to lack of adaptation of the recommended pediatric dosage to an adolescent. The fourteen-year-old girl was admitted to hospital for fever and confusion. Herpetic encephalitis was quickly diagnosed on the basis of the clinical picture of loss of consciousness, intra-cranial hypertension and localized neurological signs, lymphocytic meningitis and right temporal edema. The diagnosis was confirmed by PCR. Treatment comprised restriction of fluids and intravenous infusion of acyclovir (500 mg/m^2/8 h). Although diuresis and blood electrolytes remained normal, from the 4th day creatinemia rose from 62 µmol/l to 192 µmol/l. On the 6th day creatinine clearance was 20 ml/mn/1.73 m^2. Doppler echography of the kidneys was normal and blood levels of aciclovir ten hours after infusion were 40 µmol/l (day 5). The treatment was withdrawn for 24 hours and recommenced at a dose of 10 mg/kg/8 h by slow infusion over 2 hours with normal fluid intake. Renal function normalized in one week. Acute renal failure during acyclovir treatment is secondary to obstructive renal disease caused by crystallization of the acyclovir in the tubule. Crystalluria is dose-dependent, renal failure usually occurring with plasma concentrations of acyclovir higher than 20 µg/ml, explaining why renal failure mostly occurs in two circumstances : with intra-venous administration (especially in bolus form) and in dehydrated patients or those with renal disease (the reduced urine flow further reduces the ability to dissolve the acyclovir). In this young person the fluid restriction and the high dose led to the occurrence of kidney failure. Finally, it is necessary to adapt the dose recommended for children and infants for adolescents as they approach "adulthood" and to survey renal function, especially in cases of fluid restriction.

P6
Recombinant Hepatitis B Vaccine: Long-term Follow up of Preterm Infant Immune Response and Comparison of Two Vaccination Protocols

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Background: A three year follow-up study of hepatitis B antibody (HbsAb) persistence in low- risk preterm infants: 3-dose protocols using recombinant hepatitis B vaccine (HBV) were compared: 1. Initial vaccination 24 hours after birth and 2. Vaccination delayed until a weight of 2000g was achieved.

Methods: 136 children of HbsAg negative mothers included: children born preterm (≤ 35 weeks, *n*=57) and children born full-term (≥ 37 weeks, *n*=39), received the first dose of HBV within 24 hours of birth; children born preterm (*n*=40), received the first dose of HBV when a weight of 2000g was reached. All infants received a 2nd dose one month after the first, and a 3rd dose 5 months later. HBsAb was measured at age 3-3.5 years (≥ 2.5 years after completion of primary series). HBsAb ≥1:10 IU/L was positive.

Results: At 3–3.5 years of age, the premature-delayed-vaccination group had a significantly higher percentage of positive HBsAb levels and a significantly higher geometric mean concentration (GMC) than the premature or full-term groups vaccinated at birth. (93% vs 54.4%, *P*<0.001 and vs 71.8%, *P*<0.05 respectively; 119 vs14.2, *P*<0.001 and vs 32.7, *P*<0.005 respectively).

Conclusions: HbsAb persisted at a significantly higher GMC and in a significantly greater percentage of delayed vaccination premature infants at 3–$3_{1/2}$ years of age than in early-vaccinated premature and full term infants. The short-term advantage of delayed HBV vaccination of preterm infants has been shown to persist for at least the first three years of life.

P7
A Novel Scheme for the Reporting of Adverse Drug Reactions in Children

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Background: The safety of medicines used in children is of considerable public interest, yet available data to monitor the safety of medicines in children is limited. We wished to raise awareness and stimulate reporting of adverse drug reactions (ADRs) in children in the Trent region.

Methods: A pilot Paediatric Regional Monitoring Centre (PRMC) has been established in the Trent region. The scheme operates as an extension of the UK's spontaneous reporting scheme, the Yellow Card Scheme run by the Medicines Control Agency and the Committee on Safety of Medicines. Proactive interventions including a monthly reminder letter and presentations to staff in the identified hospitals have been made.

Results: During the first two years of the PRMC, 367 reports were received from the Trent Region (an average of 177 reports annually). Prior to the scheme, there had been 40 reports received in one year. There were seven deaths reported.

Conclusions: The number of ADR reports from the Trent region has increased considerably in the first two years of the scheme. The results show that intensive education and promotion of ADR reporting can result in a major increase in reporting. This initiative will increase our knowledge about the safety of medicines used to treat children and so help protect public health.

P8
Incidence of Adverse Drug Reactions in Children

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Little information has been published on the incidence of adverse drug reactions (ADRs) in children. We carried out a prospective study to evaluate the incidence of ADRs necessitating admission to hospital or referral to a private paediatrician, or occurring during hospitalisation in a paediatric unit. Intensive monitoring of the admissions to the 10 paediatric departments of the Paediatric Hospital of Tours (175 beds) was performed over a period of one week. Reasons for admission, age and treatment were recorded and all children were followed up until discharge. Regular visits were made to all wards and all adverse drug reactions were recorded. All the 39 private paediatricians of the same city were asked to include all children seen in consultation in a given week and to record the number of them referred for an adverse drug reaction. 260 children were admitted to 9 paediatric departments during the week of the study. 1019 (45.8%) had received at least one drug (median 2, range 1–11). 4 children were admitted to paediatric departments for an adverse drug reaction, i.e an incidence of 15.3 per 1000 paediatric admissions (95%CI=[4.2–38.9]). Only 227 of the 260 children admitted (88%(received at least one drug (median 3, range 1–14) during hospitalisation, 6 of them developed an adverse drug reaction which did not increase the length of hospitalisation, i.e. 26.4 per 1000 children hospitalized ((95% CI=[9.7-56.6]). During the same period, 428 children attended the Accident and Emergency Department (A & E). 73 (45.8%) had received at least one drug (median 2, range 1–5). four of 428 consultations in the A & E Department were for adverse drug reactions, i.e. 9.3 per 1000 paediatric consultations (95%CI=[2.5–23.7]). 16 private paediatricians agreed to take part in the study and reported seeing 1192 children during the week of the study. 8 of them consulted for an adverse drug reaction, i.e. 6.7 per 1000 children (95%CI=[2.9–13.1]). This French study is in agreement with other studies and confirms that the incidence of adverse drug reactions is lower in children than in adults.

P9
Developmental Toxicity and Estrogenicity of UV screens

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The presence of UV screens in the environment (surface water) and the evidence for accumulation in the biosphere (fish and humans) raises the question of possible longterm effects of this diverse class of compounds. We recently demonstrated estrogenic activity *in vitro* on MCF-7 cells for five out of six frequently used UV screens (Bp-3: benzophenone-3; B-MDM: butyl-methoxydibenzoylmethane; HMS: 2-hydroxibenzoic acid–3,3,5-trimethyl-cyclohexyl ester; 4-MBC: 4-methyl- benzylidene camphor; OD-PABA: octyl-dimethyl-*P*-aminobenzoic acid; OMC: octylmethoxicinnamate;). 4-MBC, OMC and Bp-3 were also active *in vivo* by the oral route (uterotrophic assay on immature Long Evans (LE) rats)[1]. In further tests for transdermal activity, immature hairless (h/h) rats were dermally exposed twice daily for $5_{1/2}$ days by immersion of the animal up to the shoulders in olive oil containing the chemicals. 4-MBC and OMC increased uterine weight. Experiments in progress further show an increase in the number of bromo-deoxyuridine labeled nuclei in uterus, indicating increased cell proliferation. Because of the potential sensitivity of developmental processes to endocrine active chemicals, we started a developmental toxicity study on 4-MBC. The chemical is administered in the chow (0.1–1g 4-MBC/kg chow) yielding daily doses of 7.0–70 mg/kg body weight, during a minimum of 10 weeks to adult male and female LE rats of the F0 generation before mating, then during pregnancy and lactation, and to the F1 offspring until adulthood. So far we observed a dose-dependent decrease in litter size and a reduced survival rate at postnatal day (PN) 14. Surviving male rats showed a dose-dependent reduction of testes weight at PN 14 at 23–70 mg/kg/day.

Reference

1. Schlumpf M, *et al*., Environ. Health Perspect, 2001;109: 239–245.

P10
Direct and Adrenal-Mediated Effects of Nicotine on Sex Steroid Synthesis in Fetal and Neonatal Rats

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Prenatal nicotine exposure impairs sexual rat brain differentiation and reduces the gestational day (GD)18 testosterone peak[1]. We studied interactions of nicotine and its metabolite cotinine with steroid biosynthesis during sexual brain differentiation. Testosterone synthesis from [^{3}H]progesterone by testis homogenates of postnatal day (PN)2 rats (PN 1 = day of birth), was inhibited by cotinine (Ki=146 μM), but not nicotine. With [1β-3H]androstenedione as substrate, both compounds inhibited aromatase in basal forebrain homogenates of GD19 (nicotine, Ki=409 μM, cotinine, Ki=214 μM) and PN2 male rats (nicotine, Ki=400 μM). After nicotine treatment from GD12 by minipump at a dose (1.9 mg/kg/day) found to affect postnatal brain aromatase activity[2], nicotine analyzed by GC-MS, was higher in fetal (GD19) brain (0.80± 0.05 μM) than in maternal plasma (0.31±0.01 μM), while cotinine levels were similar (2.25±0.14 μM, 2.83±0.17 μM). The discrepancy between drug levels and Ki values speaks against a significant role of direct inhibition of steroid synthesis. Therefore, we tested possible indirect mechanisms. We previously observed a rise of plasma corticosterone in GD18 male rats[2] at the time of suppression of the testosterone peak. Corticosterone inhibits testicular testosterone synthesis; this action is diminished by conversion to 11-dehydrocorticosterone by 11β-hydroxysteroid dehydrogenase. Inhibition of this enzyme by metyrapone (150 mg/kg, GD17) restored testosterone in nicotine-exposed fetuses to the control level at GD18 (Control 1.64±0.13 nM, nicotine 0.84±0.18 nM, nicotine+ metyrapone 1.54±0.17nM, P<0.01). This suggests that the inhibition of fetal testosterone secretion by nicotine is mediated by corticosteroids.

Reference

1. Lichtensteiger W, and Schlumpf M, Dev. Brain Dysfunct 1993; 6: 279
2. Von Ziegler NI, Schlumpf M, Lichtensteiger W, Dev. Brain Res 1991; 62: 23.

P11
Expression of Cyclooxygenase-1 and Prostaglandin Receptors in Growth Plate and Articular Cartilage Of The Rat: an *In Situ* Study

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Background: An important role of Prostaglandin E_2 (PGE_2) in growth plate chondrocyte metabolism was demonstrated by *in vitro* studies. The effects of PGE_2 are mediated by the receptors EP-1 to EP-4. However, expression of EP receptors and COX-1 were not analysed in growth plate and articular cartilage *in situ* to confirm their physiological and pathophysiological role *in vivo*.

Methods: Frozen sections (4μm) from the rat proximal tibia were fixed in paraformaldehyde (4%) and stained by the alkaline-phosphatase-anti-alkaline-phosphatase (APAAP) method. Polyclonal rabbit antibodies against COX-1 (1:100), EP-1 (1:200) and EP-2 (1:200) as well as collagen II (1:100) and collagen X (1:200) were used. For detection of gene expression, RT-PCR was performed on mRNA from homogenised growth plate cartilage.

Results: Immunohistochemistry showed a homogenous expression of EP-1 and EP-2 receptor in all differentiation zones of the growth plate. This was paralleled by detection of EP-1 and EP-2 mRNA by RT-PCR. In addition positive reaction for COX-1 could be demonstrated. In contrast, articular cartilage chondrocytes elucidated the expression of EP-1, EP-2 and COX-1 in a markedly different pattern with less expression in apical cell layers. Collagen II, localised in all growth plate and articular chondrocytes as well as Collagen X, solely expressed in the hypertrophic zone of growth plate, functioned as positive control.

Conclusions: In our study we demonstrated the *in situ* expression of EP-1 and EP-2 as well as COX-1. These findings warrant further studies on the physiological and pathophysiological relevance of the prostaglandin system in chondrocytes.

P12
Therapeutic Drug Monitoring of Gentamicin in Serum and Saliva of Children: Once-Daily as Compared to Twice or Thrice-Daily

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Background: Gentamicin is widely used in pediatric medicine, and therapeutic monitoring is mandatory due to the narrow margin of safety and wide inter and intra-patient pharmacokinetic variabilities. Saliva sampling may be of potential interest, especially in children, in whom blood sampling is often difficult. Experience with once daily intravenous administration of aminoglycosides has grown in recent years, mainly in adults.

Methods: In this study, gentamicin levels were measured in serum and saliva of 39 children treated for urinary tract infection. 12 patients received gentamicin three times a day, 12 received the drug twice daily and 15 once-daily.

Results: Trough serum gentamicin level was lower among children receiving the drug once-daily as compared to those receiving it twice or trice daily, but not statistically so. No correlation was found between serum gentamicin levels and saliva levels when the drug was administered three times or twice a day; however, a good correlation was found when the drug was administered once-daily (r^2=0.89, P<0.0001).

Conclusions: Saliva sampling may be used as a noninvasive method of measuring gentamicin serum levels to guide dosage adjustments in patients receiving the drug once-daily.

P13
Microdialysis in Venous Blood Samples Adding a Colloid to the Perfusion Fluid – an *In Vitro* Study

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Background: Pharmacokinetic investigations are mandatory in children of different ages to ensure adequate dosing. However, blood sampling in newborns and infants is limited due to small blood volumes. Microdialysis offers a technique to quantitate unbound drug concentrations in plasma without removing any blood cells out of the body. Therefore, we developed a microdialysis technique to quantitate the B-receptor blocker sotalol in blood of children who suffer from supraventricular tachycardia (SVT).

Methods: *In vitro* experiments were performed to optimize recovery of sotalol in the perfusion fluid (Ringers solution in mmol/l: sodium 147, potassium 4, calcium 2.25, chloride 155). Microdialysis catheters were immersed in Ringers solution, in citrate plasma and in heparinized whole blood containing sotalol concentrations which can be expected in vivo (2.30, ,1.15 0.58,, 0.23 and 0.12 μg/ml). Perfusion flow rates were: 0.3, 0.5, 1.0, 2.0, and 5.0μl/min. Changes in colloid osmotic pressure were induced by using perfusate with and without addition of 40g/l dextran- 70.

Results: In Ringers solution, plasma and blood, the entire sotalol concentration range could be detected in the perfusion fluid at all flow rates. However, sotalol recovery was 100.2 ±6.7% (n=5) at flow rates of 1.0 l/min in Ringers solution but dropped to only 76.4 ±11.6% (n=5) in plasma and 74.8 ±24.5% (n=4) in whole blood. Adding 40g/l dextran-70 to the perfusion fluid restored recovery of sotalol to 97.9 ±15.5% (n=8), indicating that besides isotonic conditions, adequate colloid osmotic conditions are essential to ensure complete equilibration between blood and perfusion fluid at low flow rates. (Values are mean±SD).

Conclusion: *In vitro* experiments showed that the micordialysis technique is applicable to measure sotalol in blood and plasma in concentrations which can be expected during sotalol therapy in children with SVTs. Adding dextran to the perfusion fluid ensures complete equilibration between blood and perfusion fluid and thus enables quantification of sotalol for *in vivo* experiments in future.

This study is supported by the German Heart Foundation.

P14
Unusual Side Effect Following Anti-Hemophilus Vaccination: Acute Edematous Reaction

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Clinical trials prior to release of pentavalent vaccines (DTPPHib) failed to reveal any specific side effect, and the pattern of tolerance was similar to that of the available tetravalent (DTPP) vaccines. Following several years of use, a new side effect in the form of an acute edematous reaction has been reported with vaccines containing the Hib component. We report an analysis of cases of acute edematous reactions associated with this vaccine and reported to the French Regional Drug Monitoring Centers. The cases reported involved edema of the lower limbs, sometimes associated with cyanosis or transient purpura, affecting the whole vaccinated limb and sometimes also the contralateral limb. The reaction, sometimes accompanied by fever, pain and weeping, occurred in the hours following vaccination and lasted for several hours, disappearing spontaneously and without sequelae. It occurred in most cases after the first injection and did not recur if DTPP and Hib were injected at separate sites and on different days. Its incidence has been evaluated at between 0.95 and 2.2/100,000 children vaccinated. Acute edematous reactions have not, to our knowledge, been reported with vaccines containing isolated or combined DTPP components. They appear, therefore, to relate to the Hib component and to be more frequent if the vaccine is administered in combination. These edematous reactions do not correspond to any known clinical entity. There are three hypotheses to explain the mechanism involved: 1) hypersensitivity vasculitis, similar to acute infantile hemorrhagic edema; 2) type I allergic reaction demonstrated by a deep urticarial reaction (angio-edema?); 3) capillary leak syndrome. Such acute edematous reactions do not modify the benefit:risk ratio of anti-hemophilus b vaccination, but they must be recognized by physicians when decisions are made concerning subsequent injections (separate dates and sites of injection of Hib and DTPP components).

P15
Amphotericin B Colloidal Dispersion Use in Very Low Birth Weight Infants with Poor Renal Function

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Background: Open label study: Invasive Candida infections in premature infants treated with amphotericin B colloidal dispersion (ABCD).

Methods: Hospitalized premature infants, with a blood or urine culture positive for Candida sp. and serum creatinine ≥ 1.2mg/dl were eligible. Daily tests included: renal, hepatic function; blood counts; blood cultures until three consecutive cultures were negative. Serum creatinine ≥ 1.2 mg/dl at end of therapy required a 6 month follow up. ABCD dosage: 3 mg/kg/day IV on day 1 and 5 mg/kg/day IV thereafter. Positive cultures for more than 7 days required addition of a second antifungal drug.

Results: 16 preterm infants (779 ± 170g, 25 ± 2 weeks) met the study criteria:15 had positive blood cultures; 1 had a positive urine culture only. Two infants died within 24 hours of the first dose, of causes unrelated to treatment. Two infants died during treatment. One died of pneumothorax on day 5 of treatment. The other had Candida negative blood cultures from day 12, but was treated with ABCD and fluconazole for a suspected atrial fungal ball until death from bacterial sepsis with multisystem failure on day 50, ABCD treatment led to a cure in 12/14 cases: as a sole agent in 8 (57%), and with a second agent in 5 (36%). No acute events were related to ABCD infusion. Normal renal function returned in 10/12 surviving infants by end of treatment and in 1/12 within six months. No side effects were found.

Conclusion: ABCD effectively treated 10/12 evaluable cases of candidiasis in ELBW infants, without drug-related side effects. We suggest a large scale randomized double-blind study of ABCD versus conventional amphotericin B in premature infants to confirm these findings.

P16
Intrathecal drug delivery in children with brain tumours

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Background: Intrathecal drug therapy has been shown to be successful in treating the leptomeningeal spread of malignancies such as leukaemia and lymphoma. It has rarely been explored as a treatment for primary CNS tumours. Treatment is currently surgical resection and radiation. Radiation causes neuro-cognitive sequelae affecting the quality of life of survivors. Alternative means of treating these patients to improve survival rates and reduce toxicity is urgently needed. Intrathecal drug delivery may offer this alternative by bypassing the blood brain-barrier and allowing cytotoxic drugs access to tumour cells. The aim of this project was to identify drugs suitable for further investigation for intrathecal administration based on their physicochemical characteristics and current knowledge of use.

Methods: Available chemotherapy agents were identified from pharmaceutical textbooks. Literature searches using Medline and Embase were conducted to identify relevant physicochemical, efficacy and toxicity data on all drugs.

Results: 30,033 citations were screened covering 111 cytotoxic drugs. 90 drugs were excluded for a variety of reasons. The remainder were prioritised in terms of their potential usefulness for intrathecal administration to children with brain tumours.

Conclusion: Drugs showing potential for administration by the intrathecal route in children with brain tumours were identified. Further work should determine which drugs will be worthy of testing in clinical trials.

P17
The Safety of Metoclopramide for Nausea and Vomiting of Pregnancy

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Background: Nausea and vomiting are very common during pregnancy, mainly throughout the first trimester. The cause of nausea and vomiting during pregnancy is yet unknown. Most cases of nausea and vomiting in pregnancy are mild and do not require any specific therapy. Nevertheless, in some cases drug treatment is needed. Metoclopramide is a dopamine receptor blocking drug that is used to treat various gastrointestinal conditions, and was found to be a more effective treatment of nausea and vomiting than placebo.

Objective: The aim of this prospective study was to investigate the effect on the fetus of intrauterine exposure to metoclopramide in terms of teratogenicity.

Methods: Women who received metoclopramide and consulted five teratogen information centers in Israel, Italy and Brazil were studied. Cases were paired for age, smoking and alcohol consumption with controls exposed to nonteratogens.

Results: 126 pregnant women were exposed to metoclopramide during the first trimester, at a dose of 23±10 (10–40) mg for 10±10 (1–35) days. There were no differences in maternal history, birth weight, gestational age at delivery, rates of live births, spontaneous or therapeutic abortions, fetal distress and either major or minor malformations among groups.

Conclusions: The use of metoclopramide during the first trimester of pregnancy does not appear to be associated with an increased risk of malformations, spontaneous abortions or decreased birth weight.

P18
Allergy to Beta-Lactam Antibiotics in Children in Southern Israel

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Background: A history of allergy to beta lactams is poorly predictive of subsequent allergic reactions to these drugs. Skin testing (to major and minor determinants) complemented by oral challenge, is safe and highly predictive to assess a type I reaction to these drugs.

Objectives: Determine (a) prevalence of history of beta lactam allergy in children Southern Israel; (b) beta- lactam allergy in children with reported allergic reactions to beta- lactams by skin tests and challenge; (c) re-sensitization to the skin test or challenge by a second battery of tests.

Methods: (1) Medical records review. Medical records in primary clinics of Jewish (*n*= 11,069) and Bedouin (*n*= 15,586) children were reviewed. (2) Allergy tests. Children suspected to be allergic to a beta-lactam underwent skin testing (prick and intradermal) and oral challenge. Negative skin tests were followed by oral challenge with the relevant antibiotic. Reagents used were benzyl-penicilloyl-polylysine ("Pre-pen", major determinant), benzylpenicillin, minor determinant mixtures of penicillin, ampicillin, cloxacillin and cephalosporins, and normal saline and histamine (controls). Possibility of re-sensitization caused by the first set of tests was studied by repeated skin and challenge tests 1-5 months later.

Results: (1) Medical records review: 344 children (1.3%) had diagnosis of allergy to one or more beta-lactams (2.1% Jewish and 0.8% Bedouin, $P < 0.001$). Boys/ girls ratio was 1.7 ($P<0.01$). Beta lactams implicated were amoxycillin (41.9%), penicillin (28.8%), cephalosporins (21.5%), "Augmentin" (14.2%), and more than one (9.5%). Allergic reactions reported were rash (47.1%), urticaria (20.3%), angioedema (3.5%), other (8.1%) and not-specified (29%). (2) Allergy tests: 71 children with history of beta-lactam allergy were tested. 43 were diagnosed as non-allergic to beta-lactams, and 5 allergic. All allergic reactions were mild and subsided without treatment. 23 children with a negative first set of tests have not concluded the second set.

Conclusions: Skin testing to beta-lactams with major and minor determinants and oral challenge are a safe and highly predictive procedure.

P19
QT Lengthening in Premature Infants Treated with Doxapram

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Background: doxapram (Dox) is a respiratory stimulant reported to be effective in recurrent apneas of prematurity (Burnard 1978). Previous studies have reported tachyarrhythmias in adults (Stephen 1966) and atrioventricular block in neonates (De Villiers 1998) associated with Dox use. The objective of this study was to assess prospectively a possible adverse effect of Dox on preterm infants'heart activity.

Methods: 40 preterm infants (GA: 28.8±0.3 weeks, BW: 1228.2±57.8 g, mean±1 ESM), poorly responsive to caffeine, were given Dox (0.5–1 mg/kg/h IV or 30 mg/kg/d orally) in association with caffeine. They were monitored for vital signs and tolerance before Dox, 24, 48 and 72 h after the onset of the treatment: arterial blood pressure, ECG, feeding troubles and jitteriness. Plasma concentrations were measured for Dox and its major active metabolite keto-Dox and kept < 4mg/l (Dox+keto-Dox). Data were analyzed by variance analysis, Chi^2 or Fisher tests when appropriate.

Results: a statistically significant lengthening in QTc has been displayed: from 394 $\pm$ 4 ms before Dox to 409 $\pm$ 4 ms at 48 and 72 h ($P = 0.0065$). In six cases, QTc became $\geq$ 450 ms, without any rhythm and conduction alterations. No significant increase was observed in blood pressure at any time. Digestive intolerance was observed in 20 cases, associated with septicemia in 9 cases. No relationship was established with the drug plasma concentrations kept within the therapeutic range.

Conclusion: this study evidences a risk of QTc lengthening related to Dox administration associated with caffeine in preterm neonates. That should underline the interest of making an ECG survey before and during treatment. The close monitoring of plasma concentrations may be also beneficial since we have not observed any cardiovascular adverse effects although a long experience of Dox use in very low birth weight infants.

P20
A Transdermal Formulation of Paracetamol: *In Vitro* and *In Vivo* Preclinical Studies

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Background: Paracetamol (APAP) is one of the most widely used drugs in infants and children. Available formulations for oral and rectal administration of APAP may not be practical in young patients with vomiting and diarrhea and in those who refuse to take the full dose of the medication. Thus, an alternative route of administration may be most useful for pediatric use.

Purpose: Preliminary pre- clinical *in-vitro* and *in-vivo* kinetic and toxicity studies of a novel transdermal APAP formulation.

Methods: (1) Preparation of a transdermal patch containing paracetamol in an hydrogel medium. (2) *In vitro* APAP penetration through hairless mouse skin using a Franz diffusion cell system. (3) Plasma APAP pharmacokinetics following application of patches (1.8 cm^2) on depilated abdominal skin of adult rats. Six rats were treated with regular 100 mg APAP patch and 6 rats with 100 mg APAP in an enhancer- containing transdermal system. Blood samples were taken at 0, 1, 2, 3, 4, 5, 7 and 9 hours from the time of application. APAP was measured using EMIT and HPLC techniques. (4) Local (skin) and systemic toxicity.

Results: After 8 hours of application the drug remaining in the patches was 46.9±7.3% (mean±SD) of the initial dose. When a penetration enhancer was added into the formulation texture, the extent of APAP penetration was increased 25 fold. The permeability coefficient (Kp, Fick's equation) raised from 0.28×10^{-3} (without enhancer) to 12.3×10^{-3} (with enhancer). This dramatic increase in APAP permeability was much greater than the penetration following dermal application of pure alcoholic solutions (Kp 2.5×10^{-3}). The pharmacokinetic data supported the *in-vitro* results with increased penetration using an enhancer and reaching therapeutic plasma concentrations. No local or systemic toxicity was noted.

Conclusion: These preliminary data supports the concept that a transdermal APAP drug delivery system may be feasible for clinical use. Transdermal APAP penetration may be controlled by tuning the enhancer's concentration.

P21
Drug Utilization–Problem of Pregnancy

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Objective: Consumption of drugs during pregnancy is considered as a specific medical problem. Drug consumption and drug compliance in pregnancy were analysed in our study.

Methods: Structured questionnaire and data from mothers were used as principal sources for analysis. Consumption of drugs and compliance with therapy were evaluated in 61 pregnant women, 60% city and 40% rural population, of Region Nitra, Slovakia, after their child birth.

Results: Drug utilization was confirmed in 75% pregnant women. Consumption was significantly higher in the first and second pregnancies in comparison to the third or higher pregnancies. Medical prescribing was the reason of drug use in 89% pregnant women. The number of prescription drugs was higher in the first and second pregnancies. We noted medical prescribing drug compliance in 70% pregnant women. The total utilisation of analgesic drugs was 15%.

Conclusion: Adverse drug reactions (gastrointestinal problems and headache) were reported by 15 % pregnant women. 8% of infants were born prematurely. Two newborns developed sepsis. There was no correlation between child abnormalities and drug consumption during pregnacy (there were three minor congenital anomalies).

VOLUME 4 CONTENTS

Paediatric and Perinatal Drug Therapy

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