ARTICLE

Neuraxial Analgesia In Neonates And Infants: Review of Clinical and Preclinical Strategies for the Development of Safety and Efficacy Data

Suellen M. Walker, Tony L. Yaksh
Institute of Child Health, University College London

Abstract
Neuraxial agents provide robust pain control, have the potential to improve outcomes, and are an important component of the perioperative care of children. Opioids or clonidine improve analgesia when added to perioperative epidural infusions; analgesia is significantly prolonged by addition of clonidine, ketamine, neostigmine or tramadol to single shot caudal injections of local anesthetic; and neonatal intrathecal anesthesia/analgesia is increasing in some centers. However, it is difficult to determine the relative risk-benefit of different techniques and drugs without detailed and sensitive data related to analgesia requirements, side-effects, and follow-up. Current data related to benefits and complications in neonates and infants are summarized, but variability in current neuraxial drug use reflects the relative lack of high quality evidence. Recent preclinical reports of adverse effects of general anesthetics on the developing brain have increased awareness of the potential benefit of neuraxial anesthesia/analgesia to avoid or reduce general anesthetic dose requirements. However, the developing spinal cord is also vulnerable to drug-related toxicity, and although there are well-established preclinical models and criteria for assessing spinal cord toxicity in adult animals, until recently there had been no systematic evaluation during early life. Therefore, the second half of this review presents preclinical data evaluating age-dependent changes in the pharmacodynamic response to different spinal analgesics, and recent studies evaluating spinal toxicity in specific developmental models. Finally, we advocate use of neuraxial agents with the widest demonstrable safety margin and suggest minimum standards for preclinical evaluation prior to adoption of new analgesics or preparations into routine clinical practice.

Introduction
The consequences of inadequate regulation of pain were made evident by early clinical studies showing that anesthesia and analgesia reduced morbidity and mortality following cardiac surgery in the newborn1,2. As well as deleterious acute physiologic consequences, there is an evolving literature indicating that neonatal surgery and/or intensive care can result in prolonged changes in sensory processing3–6 and altered responses to future pain7–9. While adequate intra-operative anesthesia and analgesia in the newborn, as in the adult, can be achieved by inhalants and intravenous drugs, there has long been an appreciation of the benefits of neuraxial anesthetics and analgesics, which can create dense local anesthesia and analgesia that extends

into the perioperative period with reduced systemic side effects. The use of neuraxial drugs in the control of pain may now be further encouraged as recent data demonstrate that general anesthetics (NMDA antagonists, isoflurane, nitrous oxide) and benzodiazepines produce developmentally regulated increases in perinatal apoptosis and long term deleterious behavioral changes 10–12. However, it is important to appreciate that neuraxial delivery employs agents which until recently have never been systematically assessed for their safety during early development. This has been highlighted by the Anesthetic and Life Support Drugs Advisory Committee of the FDA*, which stated that "the potential for anesthetic agent-induced neurodegeneration at the level of the spinal cord should be evaluated, particularly with respect to the local anesthetics and opioids administered neuraxially".

An increasing number of drugs and preparations have been used to produce neuraxial analgesia, with clinical studies demonstrating tolerability and efficacy. However, high quality evidence for improved clinical outcomes, particularly in neonates and infants, is limited. There is a growing emphasis on the need for preclinical evaluation of spinal toxicity to fully evaluate the relative benefits and risks of different agents prior to clinical use. This is reflected by the adoption of specific guidelines for publication of neuraxial clinical trials by several major journals. In this review, we seek to address four specific issues: 1) summarize the clinical use of neuraxial techniques in neonates and infants; 2) highlight current difficulties in evaluating the comparative benefit and potential risk of different spinal analgesic drugs; 3) summarize preclinical models evaluating developmental changes in the pharmacodynamic response to spinal analgesic drugs; and 4) review minimal standards for implementation of spinal agents in neonates to permit informed assessment between different agents in terms of efficacy and toxicity in the neonate. The review will consider agents that block conduction (i.e. local anesthetics), but will focus on those that specifically attenuate the spinal processing of pain information when administered by the intrathecal or epidural/caudal route (i.e. spinal analgesics, also often termed spinal adjuvants).

Clinical use of neuraxial analgesia and anesthesia in neonates and infants

Neuraxial delivery

The control of afferent traffic through neuraxial interventions (epidural or intrathecal delivery) can be utilized in neonates and infants as (i) a sole neuraxial anesthetic technique for abdominal and lower limb surgery 13,14; or (ii) as a supplement to reduce intraoperative general anesthetic requirements and manage peri-operative pain15,16.

Intrathecal delivery of local anesthetic produces "spinal" anesthesia. Use of neonatal spinal anesthesia is increasing in some centers17,18, with large series reporting safe and effective anesthesia and analgesia13,19,20, including use in high risk and extremely low birth weight neonates21. "Single shot" spinal anesthesia provides an alternative to general anesthesia for lower abdominal or inguinal surgery, however the clinical utility of this technique is limited by the duration of action of intrathecal local anesthetics in neonates, and conversion to general anesthesia is often required if surgical duration exceeds one hour13,19. Various techniques have been utilized in infants and neonates to prolong the duration of intrathecal anesthesia including: (i) repeat administration via an intrathecal catheter22; (ii) a combined spinal and epidural catheter (CSE) technique for upper abdominal surgery23,24; (iii) additional local anesthetic administration by the surgeon during myelomeningocoele surgery; and (iv) addition of spinal analgesic adjuvants such as opioids22,25 or clonidine26,27.

Epidural analgesia can also be used as a sole technique 28,29 or as a supplement to general anesthesia for perioperative analgesia 30 for neonatal and infant surgery. Single bolus administration 15, or infusion via a catheter advanced from the caudal space 31 or inserted at an intervertebral level in the thoracic or lumbar spine 32 is possible in even the smallest preterm

neonate 33. A range of spinal analgesics are now administered, often in conjunction with local anesthetics, with the aim of: (i) improving analgesia; (ii) reducing local anesthetic requirements and associated side-effects such as motor block; and (iii) prolonging analgesia following single shot administration.

Epidemiology

Neuraxial analgesia is used in children of all ages, but the pattern of use and choice of technique varies with age of the child, across institutions, and with time in some centers18,34. In a 1994 survey of regional anesthesia by the French-language Society of Pediatric Anesthesiologists (ADARPEF), neonates comprised 3.3% of the pediatric population, but received 3.4% of caudals, 1.8% of epidurals, and 10.9% of all spinal anesthetics.34 A similar survey in 2006 found a decrease in the use of caudals, but increased use of epidural catheters and single shot spinal anesthetics, and a greater proportion of central blocks were being performed at younger ages (5.6 vs 3.4% in neonates; 30 vs 16.5% in infants < 6 months).17 In one French center, the overall proportion of neuraxial blocks decreased from 1989 to 2005, but spinal anesthetics in neonates had become the most frequent technique, comprising 30% of the total18. The number of epidurals performed annually in UK children was stable from 2002 to 2005, with 5% of the total 10633 epidural performed in neonates and 16% in children aged between 1 month and 1 year 35.

Clinical benefits and risks of neuraxial analgesia

Potential advantages of neuraxial route in neonates and infants

In addition to minimizing the potential exposure of the developing brain to general anesthetics, neuraxial analgesia may improve postoperative outcomes for high-risk neonates who are susceptible to respiratory complications (e.g. preterm born neonates with lung disease and postoperative apnea) 36 or who require major surgery for correction of congenital anomalies 37–40. However, the magnitude of benefit of intra-operative or peri-operative neuraxial anesthesia is difficult to determine from case reports or series 41. Even in older children undergoing scoliosis surgery, meta-analysis demonstrated improved analgesia with epidural local anesthetic and opioid versus systemic opioid for adolescents, but there was insufficient data to confirm any change in respiratory outcomes, length of hospital stay, or mortality 42. In younger children, variability in study design (type of surgery; neuraxial anesthesia regimes with local anesthetic in varying concentrations and doses and different types and doses of spinal analgesic) makes systematic analysis of outcomes even more difficult. Reported benefits of neuraxial anesthesia in studies that include neonates and infants are:

Reduction in respiratory complications

i. post-operative apnea. Analysis of four trials comparing spinal and general anesthesia in neonates born preterm undergoing inguinal herniorrhaphy found a reduction in the incidence of postoperative apnea only if systemic sedatives were avoided 36. Neuraxial anesthesia and avoidance of opioids may have added advantages in neonates with central hypoventilation syndromes 43. It has been suggested that spinal anesthesia can reduce costs related to postoperative monitoring and hospitalization 44.
ii. post-operative mechanical ventilation. In a randomized trial of infants undergoing cardiac surgery, caudal morphine and local anesthetic provided some analgesic benefits over systemic morphine, but the study had insufficient power to evaluate effects on early extubation 45. In case series comparing perioperative neuraxial anesthesia with systemic opioid analgesia, the proportion of neonates requiring postoperative mechanical ventilation was reduced following gastrochisis repair 38, lung resection for congenital lung lesions 46, and Nissen fundoplication 47. Cases of improved respiratory function following major neonatal thoracic surgery with epidural analgesia have been reported 48,49.

Attenuation of stress response
Circulating levels of stress hormones such as cortisol 50,51, adrenaline and noradrenaline 22,52 are reduced when supplementary neuraxial anesthesia is added to general anesthesia.

Cardiac stability
Maintenance of cardiovascular stability has been demonstrated during neuraxial techniques in neonates 53, including CSE anesthesia for upper abdominal surgery 24 and in high-risk neonates and infants with congenital cardiac disease 54. While these observations support the safety of the technique, improved outcomes in comparisons with general anesthesia have not been confirmed.

Reduction in hospital stay
In uncontrolled trials, epidural rather than systemic analgesia reduced hospital stay following ligation of patent ductus arteriosus in infants 55 and fundoplication 47.

Improved surgical outcome
Wound dehiscence following bladder exstrophy repair in neonates was avoided with prolonged neuraxial anesthesia (mean 15 days) and sedation, but there was no comparison with other analgesic techniques 37.

Potential disadvantages of neuraxial route in neonates and infants
Complication Rates
Although severe complications following pediatric neuraxial techniques are rare, the incidence is higher in neonates and infants: 0.4% vs 0.1% for all neuraxial blocks 17 and 1.1% vs 0.49% for epidural blocks alone 35. Outcomes may be worse in neonates 56,57, with complication rates as high as 4:1000 (including 3 deaths) 56 initially reported, but more recent surveys report complication rates of 0.29% (95%CI: 0.21–0.43) for central blocks (caudal, epidural and spinal; n=10,556) 17. Following peri-operative epidural infusions (n=10,633), the rate of serious incidents approximated 0.5:1000, with an additional 0.75:1000 incidents graded as moderate severity 35.

The clinical practice setting, resource availability, and experience of individual practitioners can have a major impact on the relative risk and benefit of neuraxial anesthesia. The lack of intensive care facilities in some practice settings will increase the potential benefit of neuraxial techniques that reduce the requirement for postoperative mechanical ventilation. Management by experienced practitioners may minimize the incidence and severity of adverse events in neonates, as skilled intraoperative resuscitation was required following dural puncture or intravascular injection 34. Complications related to the use of wrong equipment (eg. inappropriate or oversized needles, excessive length of catheter introduced into space) were reported in early series 34. Pump programming or prescription errors were more common in young children (0.3% in children under 1 year versus 0.07% in 1–8 year-olds) 35. All were corrected before harm occurred, but this emphasizes the need for adequate monitoring and follow-up of patients with epidural infusions

Infection
Asymptomatic colonization of epidural catheters is common (35%) but in series of 210 58 or 1458 children 59, no local or systemic infections were reported. Age was not a clear factor, although the rate of colonization was higher for caudal than lumbar catheters in the under 3 year age group 58. In a national audit of 10,633 perioperative epidural infusions, there were 25 cases of local skin infection (ages not reported); epidural abscess was reported in 2 cases (including one infant), and an additional 16 year old patient developed signs of meningism 35. In a single center over 17 years, epidural catheter related infection, limited to the paraspinal or subcutaneous tissue,

occurred in 6 of 10,437 (0.06%) cases, including one neonate and one infant 60. All presented with back pain, pyrexia, and cellulitis; 5 also had pus visible at the catheter exit site; 3 required surgical drainage; and all recovered without neurological sequelae. Epidural catheters inserted for longer periods for chronic pain management were associated with higher rates of infection (3.2% vs 0.06%) 60.

Neurological injury

Rates of neurological injury following neuraxial analgesia range from 0.13 to 0.4 per 1000 in large series, with higher rates following epidural catheter techniques than single shot caudals. Transient neuropathy was reported following 2 per 15,013 central blocks34 and 6 per 10,633 epidural anesthetic infusions35. In addition, following a programming error that rapidly delivered 15ml of solution, a 4-month-old preterm born infant developed cauda equina syndrome with persisting neurological deficit one year later35. Suspected nerve injuries occurred following 1 of 364 thoracic, 2 of 1183 lumbar, and 1 of 8493 caudal epidural blocks, with no reported long-term deficits, and children were aged 8 years and above17. Isolated cases of neurological deficit following neuraxial anesthesia of varying severity have been reported in neonates61 and older children62–65. The relative contributions of needle trauma, surgical injury, or potential drug-related toxicity to neurological injury are difficult to determine. No neurologic sequelae were reported in a retrospective review of 750 children (52% of whom were infants) requiring cardiac surgery and treated with peri-operative epidural local anesthetic, opioid and/or clonidine 66. However, as in many studies, the duration of follow-up and the nature and sensitivity of neurological evaluation was not reported. The rates of complications may be under-estimated, particularly in young children 67, who cannot report sensory symptoms and subtle motor changes are difficult to detect in infants not yet walking. More thorough follow-up of patients following neuraxial blocks has been advocated 68.

Clinical choice of a spinal analgesic: efficacy

Local anesthetics

The primary drugs delivered neuraxially in neonates are local anesthetics and examples of the range of preparations used in neonates and infants are included in Table 1. Issues of safety with neuraxially administered local anesthetics have tended to focus on systemic toxicity and high plasma concentrations that precipitate neurological and cardiovascular complications (i.e. convulsions and arrhythmias) 69,70. Age-related alterations in pharmacokinetics result in higher free drug concentration following a bolus and accumulation of local anesthetic during infusion in neonates 71–75. As a result, infusion duration tends to be limited in the youngest patients. In a recent study of neonates following bladder exstrophy repair, epidural lidocaine was infused for an average of 15 days (range 4–30 days), but with regular monitoring of plasma lidocaine concentration 37. As will be reviewed below, it should be emphasized that although widely employed, there have until recently been no systematic studies as to potential adverse effects upon the developing spinal cord 76, and no comparative studies of different local anesthetics.

Spinal analgesics and clinical study design

Few studies have directly compared the efficacy of different spinal analgesic drugs in children of different ages. This, and the lack of systematic safety data (discussed below), makes it difficult for practitioners to make an evidence-based choice between different drugs, thus contributing to the wide variability in current clinical practice 77,78.

Evaluating data from current controlled trials is hampered by variation in methodology, particularly in the sensitivity of the outcome measures and end-points used to measure the duration and efficacy of analgesia. In neonates and infants, sample sizes are frequently small 79,80 as recruitment of large homogeneous samples is difficult, and may be further constrained by ethical

issues 81. Additional variability in the type, sensitivity and specificity of pain assessment tools utilized 78 may further reduce the power of the study.

Prolongation of analgesia

If analgesia is being titrated against individual requirements, differences in pain scores should not be seen, and therefore differences in the duration of analgesia or supplemental analgesia requirements are often used as outcome measures. The most frequent comparison is between the same dose of local anesthetic with or without a spinal analgesic, and relatively few studies evaluate the ability of spinal analgesics to reduce the required concentration of local anesthetic 82,83 or the impact of different doses of local anesthetic 84. Time to first analgesia will be influenced by: the sensitivity, frequency and inter-rater reliability of pain assessment (particularly following discharge when reliance is placed on parental interventions); the trigger for administration; and the type of supplemental analgesic. Meta-analyses have demonstrated statistically significant prolongation of analgesia with caudal clonidine 79,85,86 and ketamine 84,87. The remaining question is whether the degree of change is clinically, as well as statistically, significant. As reported increases in duration range from 2.3 to 5.3 hours, analgesia may be receding soon after the patient leaves the PACU or when ambulatory patients are leaving the hospital, and this needs to be considered when providing instructions to ward staff and parents regarding supplemental analgesia.

Supplemental analgesia

The clinical significance of a reduction in supplemental analgesia as an outcome depends on the total dose, side-effect profile and relative risk of the different treatments. A reduction in opioid requirement with addition of spinal analgesics88 has the potential to reduce opioid-related side-effects such as nausea and vomiting. However, many pediatric studies have been conducted following day case surgery, where postoperative pain scores and/or analgesic requirements are low, making it difficult to demonstrate a difference between two active treatments89. A reduction in the use of mild analgesics such as acetaminophen or NSAIDs84,87 provides evidence of an analgesic effect, but the relative risk of the spinal adjuvant must be weighed against that of the additional supplemental analgesia. We would question whether avoiding one or two doses of acetaminophen over a 24-hour period justifies the risk of neuraxial administration of a drug that has not been evaluated for spinal toxicity. In addition, studies may report only the proportion of children requiring analgesia, or the total number of doses in the whole treatment group, and therefore dose requirements and relative benefits or risks for individual patients cannot be assessed.

Route of administration

Neuraxial analgesic administration has the potential to produce analgesia at doses lower than required with systemic administration, thus reducing side-effects. Epidural morphine (12–50mcg/kg) improves analgesia 90–92, and although early systemic absorption was detected, analgesia was evident 1 and 3 hours later when plasma levels were lower than required for a systemic analgesic effect 88. Lower doses (2–5μg/kg) are effective intrathecally 93–95. The degree of dose sparing depends on the chemical properties of the drug, and for more lipophilic opioids such as fentanyl, the difference between equi-effective intrathecal, epidural and systemic doses may be less96. Minimal dose sparing has also been demonstrated with ketamine, as 0.5 to 1mg/kg is utilized in caudal studies 97,98 and the same dose systemically provides procedural sedation and analgesia 99–101, albeit for a shorter duration 102. Similarly, analgesia was prolonged when comparing caudal and intravenous administration of 2mg/kg tramadol 103. Clonidine via the intrathecal 104 or caudal 105 route has a greater effect on analgesic duration than the same dose intravenously, but effects on general anesthetic requirements and early post-operative sedation are seen with neuraxial and systemic administration.

Addition of caudal adjuvants following unilateral hernia repair in children often aims to reduce local anesthetic requirements and associated motor block, but less invasive techniques such as local infiltration and ilioinguinal block are also effective in the early postoperative period 78. Few studies have directly compared different local anesthetic techniques. When compared to dorsal penile block for circumcision, caudal bupivacaine plus ketamine was found to have no advantage 106, or to produce mild prolongation of analgesia (7.6 vs 6.2 hrs) at the cost of increased motor block 107.

Spinal analgesic drugs

In the following section we will provide a commentary on the use of analgesics that are delivered by the intrathecal or epidural/caudal route, with the aim of producing spinally-mediated analgesia (i.e. spinal analgesics or spinal adjuvant analgesic drugs), and which are typically used in conjunction with local anesthetics. Table 1 provides a systematic summary of the reported literature relevant to the several families of adjuvant analgesics. In each case, the reported dosing is provided. In many cases there is limited information related to the concentration of the different drugs within the injectate, but when co-administered with local anesthetic, the desired spread and volume of local anesthetic is often the deciding factor.

Opioids are the most frequently utilized spinal analgesics, but increased knowledge of spinal pharmacology has led to drugs such as alpha-2 adrenergic agonists (clonidine), NMDA antagonists (ketamine), GABA agonists (midazolam) and neostigmine being used alone or in combination as spinal analgesics in adults 108. Use of spinal analgesics has expanded to pediatric practice, but there is marked variability in the availability of different preparations and in the clinical use of these drugs. Surveys of pediatric anesthetists in the UK reported that 16% added clonidine, 15% ketamine and 9% epinephrine to epidural infusions 77. The proportion using clonidine as a caudal analgesic has increased (26% in 2002 and 42% in 2009), whereas use of ketamine and midazolam remained relatively constant at 32–37% and 0.5–1% respectively109,110. A survey of 25 international pediatric centers found an increase use of clonidine (18 to 23 of 25 centers) whereas use of ketamine had significantly decreased from 12 to 4 centers 111. While the majority of controlled trials have been conducted in children over 6 months of age 79, many spinal analgesics have been used in neonates and infants less than 6 months (see Table 2), despite limited evaluation of age-related changes in the pharmacodynamic profile of these drugs and no systematic evaluation of toxicity in the developing spinal cord.

Opioids

Mu opioids have been administered by epidural bolus and/or infusion and also as an intrathecal additive with local anesthetic. Morphine or fentanyl has been used most frequently in neonates and infants 22,25,30, but the use of a wide range of opioid drugs has been reported in children 6 months and older including: alfentanil 112, sufentanil 113–115, buprenorphine 116, butorphanol 117–119, diamorphine, 120,121, hydromorphone 122 and tramadol 103,123–127. In surveys of UK pediatric anesthetists, 85% used opioids for epidural analgesia77, but variability in the agent chosen (fentanyl, morphine, or diamorphine) was noted in this and an earlier survey (21% adding fentanyl and 13% adding diamorphine to caudal anesthetic blocks) 109. Although many practitioners had a minimum age for the use of epidural opioids, the cutoff varied from the neonatal period to 5 years of age 77.

Clonidine and Dexmedetomidine

Meta-analyses of caudal studies in children over 6 months of age, reported prolongation of analgesia with addition of 1–2μg/kg clonidine to local anesthetic for 2.4 (95%CI:2.6–5.5) hours

79, 3.98 (95% CI: 2.84–5.13) hours 85 and 3.68 (2.65–4.7) hours 86. Many studies reported minor sedation following clonidine, which was more severe and associated with cardiovascular side-effects at higher doses (5μg/kg) 79. Case reports of side-effects of apnea, oxygen desaturation, and bradycardia have been reported in neonates given doses of caudal clonidine (1.25–2.2 mg/kg) that are tolerated by older children 128–130. Continuous infusion of epidural clonidine 0.08–0.12 μg/kg/hr produces dose-dependent analgesia when added to local anesthetic infusions 131, and higher doses of clonidine alone (0.2μg/kg/hr preceded by bolus of 2μg/kg) provide analgesia at rest following abdominal surgery 132. When added to intrathecal local anesthetic in neonates relatively large doses of clonidine (up to 2mcg/kg) prolonged analgesia 26. A subsequent observational study with longer follow-up (24 hrs) found over half of the patients were sedated in the immediate postoperative period, and the proportion of neonates developing self-limiting apnea increased postoperatively 27. This dosing represents concentrations up to 5mcg/ml being utilized for both caudal and intrathecal single shot injections and 0.6 to 1mcg/ml for continuous epidural infusion.

The more selective alpha2-adrenergic agonist dexmedetomidine (1μg/kg) prolonged analgesia when added to caudal bupivacaine, and reduced supplemental analgesic requirements by 1–2 doses of acetaminophen 10mg/kg in the first 24 post-operative hours 133. Similar analgesia was reported when comparing caudal dexmedetomidine and clonidine in children aged 6 months and above 134. As there has been limited evaluation of neurotoxicity with this drug135, further testing is required before routine clinical use 136.

Ketamine

Caudal ketamine has been utilized for perioperative analgesia in children, including neonates and infants 84,87,97,98,137. Dose ranging studies using 0.25–1mg/kg reported 0.5mg/kg as the optimum dose, with increasing side-effects at 1mg/kg 83,138–140. Recent meta-analyses evaluating addition of ketamine to caudal local anesthetic reported prolongation of analgesia for 2.26 hours (95%CI:1.53–2.98) 87 or 5.3 (95%CI:5.45–5.76) hours 84. Acute psychomimetic effects were reported in 2 of 7 trials 84, but the difference was not statistically significant in the other analysis (OR=1.72, 95%CI:0.69–4.26) 87. A reduction in supplementary analgesics was demonstrated in studies utilizing non-opioid analgesics 87 or acetaminophen (paracetamol)84, but not in studies where peri-operative opioids were required 87. Ketamine 0.5–1mg/kg was diluted with 0.5–1.0 mls/kg of local anaesthetic or saline resulting in final concentrations approximating 0.5–1.3mcg/ml 137,139,141.

Systemically administered S-ketamine has increased potency over the racemic mixture 100. Dose sparing has not been evident in caudal studies, with s-ketamine utilized in doses of 0.5mg/kg 142,143 or 1mg/kg 102,141,144. Ketamine solutions may contain benzethonium chloride 145, but there is limited information about the injectate preparation in some studies 112,146,147, while others report using a preservative free solution of racemic 106,107,138,139,148,149 or S-ketamine 97,98,137. In some regions, the number of centers using neuraxial ketamine in children has reduced in recent years 111,150.

Midazolam

Midazolam is a GABA-A agonist with potential analgesic actions in the spinal cord, but major concerns have been raised about the safety of neuraxial administration in both adult 151,152 and pediatric practice 153. Addition of midazolam 50 μg/kg to caudal local anesthetic prolongs analgesia and increased sedation in children aged 1–12 years 154. Some reports employ a preservative-free solution 148,155, but others give no details of the pharmaceutical preparation

154,156 although one reported using a solution with a pH of 6.2 rather than 3.3–3.9 as used in previous studies 157. Solutions of 0.1–0.5% midazolam were administered with 0.5–1.0ml/kg of local anaesthetic or saline resulting in final concentrations approximating 50–100 mcg/ml 154,155,157,158.

Neostigmine

Neostigmine produces analgesia following neuraxial administration in adults 159,160, but the incidence of side-effects has led to its role in pediatric practice being questioned161. Doses of caudal neostigmine ranging from 1 to 4 μg/kg have been administered in children from 5 months of age 162–166 and prolong analgesia by 9.9hrs (95%CI: 7.8–12.2hrs) but without a clear dose-response relationship 86. The relative risk of PONV is significantly increased (RR 1.78, 95%CI: 1.11–2.85] 86, with incidences from 30% 167 and up to 60% with higher doses 168. Preparations containing methylparaben and propylparaben 148,169 and preservative free solutions 170 have been utilized. Prolongation of hyperbaric bupivacaine block has also been demonstrated with intrathecal neostigmine 0.75–1mcg/kg in infants 171. This dosing represents concentrations of 2–4mcg/ml for caudal injections and 10mcg/ml administered intrathecally.

Clinical choice of spinal analgesic: safety

For the last two decades, there has been an increasing appreciation that there needs to be a specific intent to define the safety of neuraxially delivered drugs prior to routine clinical use in adults 172,173. We, and others, have argued that systematic preclinical assessment of potential for spinal toxicity in validated models should be performed before clinical delivery into the neuraxial space of neonates and children 150,161. Without safety data, it is impossible to confirm a favorable risk-benefit ratio for neuraxial administration, or to compare the relative safety, of this wide range of drugs and preparations, and clinical trials must be undertaken with caution. So significant has become this issue, that several major journals involved in pain and anesthesia have provided specific guidelines on the acceptability of work that employs the off-label neuraxial use of novel agents, indicating that systematic preclinical safety should be available or specific FDA approval gained prior to undertaking the trial 174–177. In the following sections, we review the information that does exist regarding spinal adjuvant use in human infants; but we emphasize that in and of itself, such information does not qualify the agent being delivered as safe. Often it reflects retrospective series, limited follow-up, and the primary metric of the safety study (i.e. spinal histopathology) cannot be assessed.

Evaluation of risk

Concerns regarding the potential for toxicity following neuraxial analgesic administration have been raised in multiple reviews and editorials with calls for further preclinical testing. “It is essential to undertake extensive animal testing with further evaluation of any neurotoxic effects prior to pediatric use” 79. “Before epidural midazolam is routinely used for surgery in children, more extensive testing of its use in animals should be completed”.. and “although the extensive preclinical testing may seem burdensome, the risk-benefit relationship for epidural midazolam justifies the need” 153. Although preservatives in preparations of neostigmine 178 and ketamine 179 may contribute to potential toxicity, using a preservative-free solution does not guarantee safety. Authors reporting the use of caudal ketamine acknowledge that “as yet, no permanent neurological injury has resulted from single-shot caudal ketamine use but caution is warranted” 97, and that conclusive safety studies are required 84,100. This is particularly important as isolated cases of post-operative neurological injury have been reported in children, and neuraxial analgesia may be implicated in medicolegal claims even if other potential factors (such as peripheral compression neuropathy related to positioning) are subsequently identified 180.

It was suggested several years ago that performance of neuraxial anesthesia in healthy children required demonstration of a high therapeutic ratio and additional advantages 181. Although complications are rare 35, without information regarding tissue toxicity it is difficult to determine if the drug administered contributes to the risk. Extensive clinical use does not preclude the potential for cases of toxicity 79, as seen in adult practice with chloroprocaine 182 and lidocaine and cauda equina syndrome 183. It has also been noted that a single case of neurological injury may be sufficient to change clinical practice, bring a particular technique in general into disrepute, and thus deny many children the benefits of neuraxial analgesia 161. Therefore, further specific data comparing the efficacy and relative safety of currently available and potential new spinal analgesic agents is essential to inform clinical choice. New alternatives should only be used if improved analgesia, combined with an acceptable safety and side-effect profile, can be demonstrated 161. It should be stressed that the neuraxial route of delivery exposes local tissues (meninges, roots, spinal parenchyma) to extraordinary concentrations of agent (mg/mL), which because of local restrictions in redistribution may persist for extended intervals. Accordingly, the specific assessment of the potential toxicity of the agent must be of the highest priority. In the following sections we will review the existing preclinical data related to the safety of spinal anesthetic and analgesic agents in neonatal models.

Preclinical models of neuraxial anlagesia: developmental pharmacodynamic responses

Neonatal neuraxial delivery models

Intrathecal and epidural delivery techniques

Bolus intrathecal drugs in neonatal and infant rats can be performed with a technique similar to that described in adult mice 184. The spinal column or pelvic girdle is stabilized by one hand, and percutaneous injection is performed at the level of the cauda equina in the L5/6 interspace (rodents have 6 lumbar vertebrae) with a 30-gauge needle attached to a syringe calibrated to deliver microliter volumes. Correct placement is typically demonstrated by a tail flick on needle insertion. While it is likely that such a response represents contact with a nerve root and is a potential source of pathology 185, appropriate control studies in neonatal rats have revealed no untoward anatomic pathology related to this technique 186. Systematic training with the injection of dye and confirmation of spread within the CSF on post-mortem dissection ensures that each experimenter can consistently perform the technique 184,186. In addition, we recently used in vivo imaging following intrathecal injection of a fluorescent dye to confirm that our technique was reliable and reproducible in rat pups as young as 3 postnatal days that have an average weight around 10 grams 186.

Intrathecal catheters have been inserted via a lateral thoracic laminectomy in pups as young as P3. An injectate volume of 4µL of methylene blue produces spread from the caudal cervical to lumbar/sacral region 187, but associated motor deficits limit behavioural analysis to the contralateral limb.

Single shot percutanenous epidural injections can also be performed in rat pups, with correct epidural placement (spread along vertebral segments but lack of staining in CSF) is confirmed by co-injection of Evan's blue and post-mortem dissection 188–190.

Distribution of injectate

The distribution of the neuraxially delivered agents must be defined in any preclinical model. The volume must be adequate to deliver agent to the appropriate dermatomes used to evoke pain behavior (e.g. lumbar segments for evaluation of hindlimb withdrawal reftex sensitivity) but insufficient to acutely produce supraspinal redistribution. Recently, we confirmed that segmental spread of intrathecal dye co-varied directly with injectate volume and inversely with age in rat

pups 186. An injectate volume of 0.5 µL/g produced spread across a median of 9, 7 and 5 segments at P3, P10 and P21 respectively. Increasing the volume to 1µL/g increased spread (median number of segments 16 vs 9 at P3, 13 vs 7 at P10). This was confirmed with in vivo imaging, and larger injectate volumes of 1.5 µL/g resulted in fluorescent dye extending into the cisterna magna and supraspinal cisterns 186.

The extent of epidural spread has also been related to the volume of injectate in several species 191–193. Similarly in rat pups of different ages, injectate volumes have been based on body weight, and reflect the increasing volume of the elongating spinal canal. In neonatal rat pups, epidural administration of approximately 2 µL/gram of dye 188–190 produces spread to the mid-thoracic region following low lumbar injection.

Radioactive labeling in the spinal cord has also been used to characterize neuraxial injections. Percutaneous intrathecal injection of 2 µL in P3, or 7 µL of 3[H]-gabazine in P21 rats, produced binding throughout the thoracolumbar cord 194. Epidural injection of 3[H]morphine at P3, P10 or P21 produced similar levels of binding in the cord, all of which, as expected were much lower than levels seen following systemic administration of the same dose 189.

An important indirect assessment of correct placement is the observation of an appropriate behavioral response following injection of an analgesic or local anesthetic. While overly large volumes promoting supraspinal redistribution are to be avoided, very small volumes may in fact lead to an inadequate movement of the injectate to the spinal segments regulating the processing of afferent traffic. Accordingly, demonstration of a reliable and dose-dependent change in pain behavior is a critical component of validating dosing volumes in a preclinical model. Neuraxial local anesthetic effects may be assessed by motor and/or sensory changes, and thoracolumbar spread can be assumed by maintenance of adequate respiration, motor block restricted to the hindlimbs and/or lack of a hindlimb withdrawal response to a suprathreshold stimulus 76,186.

Developmental pharmacodynamic profile of spinal analgesics

We have postulated that evaluation of the relative safety (or toxicity) of different spinal agents is best made in the context of the therapeutic ratio i.e. the dose that produces toxicity or the maximum tolerated dose versus the dose that is required to have a therapeutic analgesic effect 186,195,196. Accordingly, it is appropriate to consider the utility of neonatal models of neuraxial delivery in defining dose-related analgesic and behavioral effects. Developmentally-regulated changes in the structure and function of nociceptor pathways, and in the expression and distribution of receptors, have a significant impact on analgesic efficacy and dose requirements during postnatal life 197. Studies in developmental models, particularly the rat pup, allow systematic assessment of a variety of specific nociceptive end-points and the degree of alteration by analgesic agents.

Analgesic efficacy and age-dependent dosing

Increases in the mechanical withdrawal threshold or thermal withdrawal latency threshold of an un-injured hindlimb can be used to evaluate age- and dose-dependent anti-nociceptive analgesic effects. The efficacy of spinal analgesics has also been evaluated by nocisponsive behaviors to local irritants such as formalin 198 or mustard oil 199, and also in facilitated hyperalgesic states such as carrageenan-induced inflammation 188,190,196.

In early life, an enhanced sensitivity to opioids is demonstrated whether given by systemic 200, epidural 189,201 or intrathecal 186,202 administration, and lower dose requirements with neuraxial administration confirm selective spinal analgesic effects 186. Changes in opioid receptor distribution in the dorsal root ganglion and spinal cord are likely to contribute, and may

also explain modality specific differences in efficacy against thermal and mechanical stimuli 189,202–204. Lower doses of opioid 186,201, local anesthetic 188, NMDA antagonist196 and alpha2 agonist190,199,205 reverse injury-induced hyperalgesia and/or increase withdrawal thresholds in neonatal rat pups when the dose is adjusted for weight.

Side effects

Effects unrelated to analgesia may be usefully considered as those which are reversible and those that are irreversible. Side-effects such as sedation, motor impairment, and cardiovascular changes can often limit dose escalation. These dose-dependent effects can be evaluated in laboratory studies and compared with analgesic doses to determine the therapeutic window (difference between dose producing side-effects and the analgesic dose) at different ages. In humans, such side effects may represent: i) a spinal action (e.g. inhibition of the micturition reflex after spinal morphine)206; ii) a direct neuraxial redistribution to the brain (as with the behavioral disruption reported after intrathecal ziconotide) 207; or iii) systemic redistribution of the neuraxial dose after intrathecal delivery (e.g. rapid sedation after intrathecal lipophillic agents such as sufentanil) 208. Side-effect end points may vary with age. Thus, in preclinical models, in addition to lower anti-hyperalgesic dose requirements with epidural dexmedetomidine, the dose that significantly prolonged the righting reflex or reduced heart rate was lower in the youngest animals, resulting in a narrower therapeutic window in early life 190,199. It should be stressed that these side effects represent adverse events that are related to the physiological and reversible pharmacodynamic profile of the particular competitive agent. Support of function, such as respiration and blood pressure, until drug clearance or reversal will often prevent any further deterioration. These events are important as they limit the useful dose range of the agent that can be practically tested and to which the patient may be safely exposed. This would be defined as the maximum tolerable dose (MTD).

In contrast, drugs at some concentration or dose exposure may exert a direct effect upon cellular function and lead to irreversible changes in cellular viability and thus represent tissue toxicity. Such end points would be, for example, expression of apoptosis or necrosis, frank demyelination, or changes in endothelial cell function. Some of these effects may be manifest by changes in spinally mediated behaviors or physiology, such as seizures, paralysis or anesthesia. On the other hand, where the tissue injury is delimited or where changes are slow and initiate compensatory actions, such effects may not be associated with functional or behavioral changes in the preclinical model. An example of this is the slowly growing, space occupying granuloma 209. Here, the appropriate criteria are the systematic post mortem assessments of target tissues (spinal cord, nerve and DRG). Without this, the absence of negative functional signs can be a false negative as regards tissue toxicity.

Preclinical spinal drugs: developmental toxicity

Impact of postnatal age

Preclinical models for assessing intrathecal and epidural drug safety have been established in adult animals 210, but there has been little effort until recently to develop models for assessing spinal toxicity throughout the early postnatal period of development. It is crucial that while persistent changes in behavior after neuraxial drug treatment maybe a signature of direct tissue toxicity, absence of such changes cannot be construed as being an absence of toxicity. Such an assertion requires demonstration of absence of neuropathology, e.g. histological signs, increases in apoptosis, and alterations in glial response in exposed tissues. We argue that an important element in considering drugs for neuraxial delivery in human neonates and infants is their appropriate preclinical evaluation. In the following sections we will consider several variables that

we believe impact upon the preclinical assessment of developmental toxicity of neuraxially delivered agents.

Activity-dependent neural development

There are well-established critical periods in early postnatal life when the normal development of neuronal circuits is activity-dependent, and alterations in neural activity can produce long-term consequences that are not seen following the same perturbation in the adult 211. Neural activity promotes synaptic strengthening and network formation; whereas lack of activity and failure to form appropriate synaptic contacts can result in programmed cell death (apoptosis). In contrast to excitotoxic cell death, apoptosis is a normal developmental process for activity-dependent matching of pre and post-synaptic populations and the refinement of neural circuits. However, during these critical periods, exposure to drugs such as general anesthetics that reduce excitation (NMDA antagonists) or enhance inhibition (GABA agonists), may trigger excessive degrees of apoptosis in many brain areas 11,212–216. The degree and distribution of apoptosis change during the first 2 postnatal weeks, with peak susceptibility in the cortex around P7 217. Prolonged general anesthesia in P7 pups increases apoptosis not only in the brain but also in the spinal cord 76,218. Changes outlined below also emphasize the significant plasticity of the developing cord. As such, neuraxially administered anesthetics and analgesics which alter neural activity in the cord may also produce specific patterns of toxicity that differ from those seen at older ages.

Developing spinal cord structure and function

During postnatal development there are significant structural and functional changes in nociceptive circuitry in the spinal cord. A-fiber afferent terminals initially project throughout the dorsal horn and only gradually withdraw to deeper laminae over the first 3 postnatal weeks in the rat as C-fiber projections mature 219,220. The normal postnatal development of A- and C-fiber innervation in the spinal cord is activity-dependent and can be altered by changing input at critical stages 221. Blockade of synaptic activity by a neuraxially administered slow-release NMDA antagonist prevents the structural re-organization of A-fiber terminals and the neonatal pattern of low mechanical withdrawal thresholds and large dorsal horn receptive fields persists into adulthood 222. The somatotopic organization of primary afferent terminal fields can also be altered by changing neural input during the neonatal period 223,224. Cell death in the DRG is a normal developmental phenomenon and is balanced by proliferation in early life 225. However, cell death occurs more rapidly and to a greater extent after sciatic nerve section in neonatal compared with adult animals 226. Importantly, responses to neonatal injury such as inflammation or surgical injury have been associated with long term functional consequences and an enhanced sensitivity to future injury 197,227–230.

The balance between excitatory and inhibitory activity in the spinal cord changes during the postnatal period197,231–233. Excitatory glutamate receptors (AMPA, NMDA and metabotropic glutamate receptors) are highly expressed and tend to be more widely distributed in the neonatal spinal cord. Developmental changes in subunit expression of the NMDA receptor are associated with changes in channel kinetics and increased calcium influx that further increase excitatory effects231,232, and may influence the potential for toxicity. GABA inhibition is functional at a cellular level, but there is minimal glycine-mediated inhibition in the neonatal spinal cord 234 and a delay in the overall maturation of inhibitory networks194,233,235,236, and local GABA-mediated inhibition in the cord is initially dominated by descending excitatory effects237,238. Ketamine and propofol have been shown to increase cell death and alter dendritic arborization of GABAergic neurons in vitro239,240 but effects in spinal networks have not been directly evaluated.

Standards for preclinical evaluation of efficacy and toxicity of spinal analgesics

Characteristics of preclinical safety evaluations

Preclinical safety evaluations by definition employ surrogate models with key characteristics that mirror those of the human condition; in this case, the mammalian neonate during the early post natal phases of development receiving spinal drug exposure in a validated model. As reviewed above, the minimal component to an appropriate assessment of toxicity is the systematic consideration of pathology in the neuraxis as compared to the appropriate neuraxial vehicle control.

Validated model and drug delivery

To date the principal developmental toxicity model employed for neuraxial delivery has been percutaneous delivery in rat pup; but the model (i.e. the animals and the delivery system) must be validated. This implies that the drug delivery has been reliably demonstrated to occur within the intrathecal space (an important issue where the delivery has been percutaneous puncture) and that the injection protocol (needle placement, volume) results in an adequate and reliable distribution of the injectate. As discussed earlier, preliminary studies are required to ensure reliability of the technique in the hands of each investigator, and to avoid confounding effects of dyes in toxicity studies correct placement can be confirmed by measuring a predetermined dose-dependent acute behavioral change (eg. increase in hindlimb withdrawal threshold or motor block). In addition, the model should have the ability and sufficient sensitivity to reveal a profile of toxicity that has been previously described (e.g. apoptosis or demyelination).

It is of fundamental importance that appropriate control groups are included to statistically differentiate between the effects of the interventions and effects of the intervention plus drug. A saline injection group will demonstrate effects related to the technique, needle trauma or volume of injectate. In addition, comparison with a naïve group ensures effects are not related to the brief anesthesia, handling or maternal separation required for the procedure 186.

Spinal toxicity in adult models has been evaluated following both epidural and intrathecal delivery. Although both intrathecal and epidural delivery have been demonstrated in the neonatal rat, current toxicity models focus on intrathecal delivery. Higher doses or concentrations of epidural drug are frequently required to achieve similar concentrations at target sites within the spinal cord. As such, the worst-case scenario is the intrathecal delivery of an intended epidural agent; not only because of the risk of increased acute side-effects, but also because of the exposure of the cord to an increased dose or concentration of drug. Cases of unrecognized dural puncture and inadvertent total spinal have been reported in large series (2 per 10,633 cases35 and 1 per 10,09817). In addition, the overall incidence of dural taps has been reported at 0.12% 16 and 0.1% (CI 95% = 0.05–0.19)17, and 6 of the ten dural taps in the latter survey were associated with caudals in babies. This further emphasizes the need to establish a safety profile for all neuraxial drugs, whether epidural or intrathecal delivery is planned.

Animal age

The infant rodent is frequently utilized as a model for evaluating the progress of postnatal mammalian development. While direct translation of different developmental ages from rodents to humans, and the specific timing of events after birth, continues to be debated, the sequence of development of sensory and reflex systems in rodents correlate with those of human infants 241. Statistical models have been developed to translate development across species 242,243, but are predominantly based on structural measures, and acknowledge that as peak synaptogenesis is more complex and more prolonged in the human, the model can not account

for activity-dependent modification following birth 244. In terms of spinal processing many approximate a P3 rat with a preterm human neonate, P7 with an infant, P21 with an adolescent, and P35 with young adulthood 197,245,246. Translational developmental models based on correlating behavioral measures support these estimates 241. In both humans and rats, locomotor capabilities develop postnatally, with a gradual rostrocaudal pattern of maturation. Rat pups ambulate through use of forelimbs and the upper torso by P3-4, crawling behavior peaks around P7, body weight is fully supported by P12-13, and rearing without foreleg support is achieved by P18 241. Spinal reflexes, which incorporate both sensory and motor development, also show similarities in the sequence of development in the postnatal rat and human infant 247,248, with gradual maturation from low threshold 190,249–251, large receptive fields 251,252, poorly directed and generalized responses 250,251,253 in both rodent and human infant early life. Clear relationships between the intensity of the stimulus and the degree of reflex withdrawal response 229,254,255 are maintained at all ages, thus facilitating evaluation of the response to injury and/or analgesia.

Vulnerability to apoptosis in the brain coincides with rapid synaptogenesis or the brain growth spurt, which occurs predominantly in the first two postnatal weeks in the rodent, but may extend from mid-gestation to several years after birth in the human infant 216. The majority of preclinical studies evaluating general anesthetic effects in the brain have focused on P7 as apoptosis peaks in the cortex at this age, and drug effects are most apparent in regions where spontaneous apoptosis is occurring 217,256. Spontaneous apoptosis occurs in the postnatal spinal cord, occurs predominantly in the dorsal horn, and peaks at a slightly earlier developmental stage than seen in the cortex with the number of apoptotic cells highest at P2-P5, and decreasing by P8-10 196,257–259. As peak apoptosis occurs at an earlier age in the spinal cord (P3 rather than P7) than the cortex, the period of susceptibility to pro-apoptotic drugs may be shorter, but prolonged general anesthesia does increase apoptosis in the cord at P7 76,218. In addition, as there are ongoing changes in the structure, function and synaptic connectivity of neural networks in the spinal cord throughout the first 3 postnatal weeks 232, assessment of developmental neuraxial toxicity should include a range of ages. This also addresses the potential uncertainty in the precise parallels between the postnatal development in the human and rodent.

Evaluation and Outcomes

This review will not seek to cover the appropriate histopathology in detail, but experts in the fields of neuropathology will argue that to define the absence of pathology, one must satisfactorily address a number of specific issues and tissue targets.

Blinded assessments

Evaluation must include an analysis that is made independent of knowledge of tissue/animal treatment, with groups that at a minimum include vehicle vs drug treatment cohorts with tissue harvested at predetermined intervals after drug exposure.

Histopathology

Analysis of pathology requires appropriate selection of histopathologial targets and indices.

i. At the minimum, it is reasonable to systematically examine hematoxylin and eosin sections to note necrosis, gliosis, and inflammation. Such examination typically includes spinal cord

and meninges, may also include dorsal root ganglia, and evaluation of nerve roots and demyelination is particularly relevant for assessing effects of local anesthetics 260,261.

ii. Evaluation of apoptosis and neuronal cell death is an important additional component in early development. Although a range of potential techniques are available 262, activated caspase-3, an enzyme in the apoptotic cascade which marks neurons progressing beyond the point of commitment to cell death 263 has been frequently used to identify apoptosis in the brain and also the spinal cord 76,186,196,218. Fluorojade C is an additional marker of neuronal degeneration 264, and we found a pattern of staining that correlated with activated caspase-3 immunohistochemistry 186,196.

iii. Activation of non-neuronal cells by the use of specific astrocyte (GFAP) and microglia (IBA1 or OX42) markers can provide further indicators of altered function and the response to injury.

iv. Evaluation of potential nerve injury requires assessment of the state of myelination. Previous work has shown that local anesthetics can produce signs of demyelination of the cauda equina 265,266. As myelin is in the developing stage up through postnatal day 3, acute effects on myelin may be difficult to assess. Others have focused on apparent changes in the root at later time points, or in the dorsal column which represents the ascending collaterals of large primary afferents 76.

v. As mechanisms associated with developmental anesthetic toxicity are further clarified, additional factors requiring evaluation in the developing spinal cord may be identified. As noted earlier, ketamine and propofol have effects on the dendritic tree of cultured cortical and hippocampal neurons.240,267,268 As changes in dendritic morphology in the spinal cord have been noted in developmental neurological disorders and have a role in synaptic plasticity after nerve injury,269,270 similar mechanisms may be relevant for developmental toxicity in the spinal cord. Neurotrophic factors and actin depolymerization have been associated with apoptosis in cultured neuronal cells exposed to propofol271 and isoflurane,272 but effects in vivo273 and relevance to analgesic toxicity in the spinal cord has not yet been established.

vi. A corollary to this commentary is that evaluation of the potential for spinal toxicity must involve the use of in-vivo animal models. Such models may be complemented by the study of drug effects in ex-vivo or in-vitro models, as has been widely employed to study local anesthetic toxicity. Changes in DRG cell function, or clonal cell viability or ex-vivo nerve exposure274–277 all provide important approaches to define potential mechanisms. However, as useful as the ex-vivo system is for characterizing local drug effects, care must be taken in extrapolating these results to the intact organism, as they can just as easily provide false positive indications which may not be relevant to in-vivo safety or pathology related to a given drug (see278).

Age at time of exposure

An important issue relates to the developmental age at initial drug exposure. As reviewed above, critical postnatal periods of neural development are represented by the onset of innervation, development of myelination of the long tract and primary afferents, and the time course of spontaneous apoptosis. On this basis, we have argued that appropriate ages in the rat are P3, P7 and P21, with P21 reflecting an animal that has essentially reached a steady state for the end points indicated

Survival time post exposure

Initiation of cell death may begin as early as 6 hours after toxin (drug) exposure, and caspase-3 immunoreactivity may be reduced at later time points as the cell decomposes 263. In the spinal cord, we found increased apoptosis 6 hours following intrathecal ketamine at P3, and significant

increases were maintained at 24 hours 196. However, glial reactions and evidence of demyelination may not be maximal until a later time point 76,196,261,266. Accordingly, an optimal characterization would include both an early (6–24 hrs) and later interval (7 day) of post treatment recovery. Longer-term effects on functional outcomes must also be considered, and be sufficiently sensitive to detect changes that are related to any observed structural or histological defects. For example, as general anesthetics at P7 increase apoptosis in the hippocampus, long-term effects on learning and memory have been evaluated 10. Although prolonged general anesthesia increased apoptosis in the spinal cord, motor performance on the Rota-rod at P30 was not altered 76,218. Whereas local anesthetic toxicity or demyelination may result in changes in motor function, spontaneous apoptosis in the ventral horn occurs mainly before birth 257. Spontaneous apoptosis 257,259 and increases following intrathecal ketamine at P3 occur predominantly in the dorsal horn, with associated long-term changes in mechanical thresholds for hindlimb withdrawal, and in static but not dynamic parameters of gait 196. This suggests that alterations in sensory and motor function should be included when evaluating long-term effects of neuraxial drugs.

Drug exposure and dose

To have credibility as a robust assessment paradigm, the drug exposure must occur at neuraxial doses which by the metric of concentration and dose equal or exceed those destined for the human condition. One limitation of percutaneous administration is that effects of dose are limited to single administrations rather than ongoing infusion and chronic exposure. Intrathecal catheterization has been reported in pups as young as P3, but motor deficits and histological damage have been noted ipsilateral to the catheter 187, thus limiting the utility of this method for assessing toxicity. The use of a single dose runs the evident risk that a drug will be observed to have pathology at a dose which is well beyond any reasonable clinical exposure. Nevertheless the higher the dose examined without pathology the more confident we can feel that the assertion of “no toxicity” is valid 173.

Translation of drug exposure and dose

An important question relates to the expression of the dosing, and the translation of dosing in the surrogate to the target species. After systemic delivery the typical metric for dose response is the body mass (eg. mg/kg). However, it is widely appreciated that across large ranges of body weight, a more appropriate metric may be body surface area, particularly when precise dosing is required to maximize the therapeutic response while minimizing the likelihood of unacceptable toxicity (e.g. chemotherapy dosing)279 (Table 3). As BSA has also been shown to correlate across mammalian species with physiological functions (such as metabolic rate, blood volume and renal function), BSA rather than body weight has been used when converting doses across species to humans. The Km factor (body weight in kg divided by BSA in m2) is often incorporated in formulae for species conversions: e.g. human equivalent dose (HED, mg/kg) = animal dose (mg/kg) × [animal Km/human Km]280. Such calculations aim to produce a comparator that generates a proportional plasma level and are important for converting no adverse effect levels (NOAELs) established in preclinical studies to doses used in clinical trials 281. However, the FDA also acknowledges that this approach has limited applicability when drugs are administered into anatomical compartments, such as the intrathecal space, where there is little subsequent distribution and where there may be as much as two fold difference in local volume 282. Considering the spinal dose in terms of mg/kg in two adult humans that may differ by a factor of two in body mass may be appropriate for avoiding systemic toxicity or side-effects associated with redistribution or inadvertent injection into vascular structures. However, variability in intrathecal volume is likely to be less, and as toxicity may be more dependent upon the compartmental volume (i.e. cerebrospinal fluid volume and/or its turn over) it is the local concentrations to which the tissue is exposed that is important 281. The problem is yet more complicated where one compares across species, and different methods for dose conversion are shown in Table 3. When

expressed as age-specific concentrations (total dose in mcg per mcl CSF volume), analgesia is achieved at twice the concentration of morphine and 42 times the concentration of ketamine and clonidine in neonatal pups. The maximum tolerated doses of intrathecal morphine 186 and clonidine 205 did not produce toxicity in the rat, despite being delivered in concentrations approximating 600 or greater than 10,000 times respectively than concentration required for clinical analgesia. By contrast, intrathecal ketamine196 produced toxicity at <150 times the clinical concentration. Although these conversions require some assumptions, and are approximate as only limited dose intervals were assessed, they provide comparisons of different agents, and again demonstrate the reduced safety margin of ketamine when compared to morphine and clonidine.

The therapeutic ratio of toxic to analgesic dose: a way forward?

The relative efficacy and safety of different treatments, and the potential benefits and risks for individual patients, are essential for choosing the most appropriate drug in clinical practice. Safety studies frequently appreciate that every agent examined neuraxially will at some point display pathology. The issue is that the drug must have a therapeutic dose that is lower than the dose that produces untoward effects upon behavior or exerts direct tissue toxicity. Ideally, this therapeutic window is wide, but depending on the desired outcome a narrower margin may be tolerated. For example, chemotherapeutic agents produce significant side-effects and toxicity, but the potential benefit for the patient is deemed to outweigh this risk. Similarly, despite concerns about pro-apoptotic effects of general anesthetics, it is clearly not appropriate to withhold anesthesia for neonates requiring surgery. However, if several agents produce a similar therapeutic end point (e.g. analgesia) what algorithm might we use to select the one least likely to have a deleterious action? One strategy is to define the therapeutic ratio of the several agents under identical conditions. In this case, one notes the quotient of the minimum dose without tissue toxicity and the minimum dose required to produce a therapeutic effect of the intrathecally delivered agent. In recent studies we showed that the therapeutic ratio in early life was > 300 for morphine and clonidine, but < 1 for ketamine as increased apoptosis occurred in the same dose range as analgesia 186,196,205. While the ratio can vary for different reasons across end points and laboratories, we would argue that in a given assessment paradigm, if two drugs have similar analgesic efficacy, but differ in their therapeutic ratio, the agent with the higher therapeutic ratio will be preferred, all other things being equal.

This particular strategy provides a rationale in the current environment to minimize the potential complications secondary to direct tissue toxicity, particularly where old drugs are being given by a new route (e.g. neuraxial) or where new drugs or preparations are being considered for neuraxial use. As noted above, with ongoing clinical use, it can become apparent that even commonly employed agents may lead to pathology; as seen following intrathecal infusion of local anesthetics 283 and chronic intrathecal morphine 284,285.

Conclusion

We acknowledge that neuraxial anesthesia is an important component of perioperative pain management in children of all ages, and particularly in neonates and infants as inadequately controlled pain in early development may also have adverse long-term effects 197. Our aim is not to discourage use of neuraxial anesthesia, but rather to encourage use of agents with demonstrated efficacy and the widest possible safety margin. Clinical studies are well suited to assessing tolerability and efficacy, but cannot reliably confirm safety and an absence of morphological effects136. Therefore, we complete this overview of neonatal neuraxial analgesic utilization by emphasizing four points.

First, we believe it is evident that the potential for spinal drug toxicity may present a greater problem in early life because of the dynamic properties intrinsic to neuraxial development.

Secondly, given the above issues, we believe that advances in this area require systematic preclinical assessments of the comparative safety of candidate agents with attention being paid to the therapeutic ratio of the neuraxially delivered agent, the developmental time of exposure to the agent, and assessment of neuropathology (apoptosis, myelination, gliosis and dendritic morphology) and long term functional outcomes. Further, the research must recognize that the critical periods of development that occur (e.g synaptogenesis, myelination and apotosis) differ for brain and spinal cord. Of equal importance, as the algorithm relating rodent and human neonatal development cannot be precisely matched, preclinical safety evaluations must review a range of developmental ages in their respective models.

Thirdly, there is a need for a greater appreciation by institutional review boards regulating clinical trials, and by editors and reviewers of scientific publications, of the issues of potential toxicity and the degree to which the clinician-investigator has adequately addressed these concerns.

Finally, we must entertain a high index of suspicion of potential toxicity when drugs are administered neuraxially. As children are rarely subject to detailed assessment after day-stay surgery, there is the potential to under-estimate the rate of complications67. This is particularly important in neonates and infants, who may not only be more susceptible to perturbations in neural development, but who are also unable to report sensory symptoms and as they are not walking, subtle motor deficits may be missed. We agree with others, that more thorough follow-up of children following neuraxial analgesia is required68, with longer-term epidemiological studies to establish clinical safety286. Integrating preclinical and clinical data has also been the focus of studies evaluating adverse neurodevelopmental outcomes following general anesthetic exposure in early life. In this situation, the clinical benefits of diagnostic investigations and surgery with adequate anesthesia outweigh the risks identified in laboratory studies, and although modifications in practice have been suggested287, current data do not support significant changes in clinical practice or provide clear evidence of a better alternative11,215. However, when considering the choice of spinal analgesic adjuvants, many provide similar analgesia but not all have undergone systematic evaluations of spinal toxicity, and changing practice to include only agents with the widest demonstrable safety margin can be achieved without compromising clinical care. It is essential to ensure that every step is taken to evaluate both the benefits and the safety of new and existing spinal drugs, prior to routine clinical use, to minimize the risk of an unexpected and untoward outcome.

Acknowledgments
Funding: Supported by: Royal College of Anaesthetists/British Journal of Anaesthesia Project Grant and Great Ormond Street Hospital Children's Charity (SMW) and NIDA 15353 (TLY)

Footnotes
*Center for Drug Evaluation and Research Anesthetic and Life Support Drugs Advisory Committee meeting on March 29, 2007. www.fda.gov/ohrms/dockets/ac/07/minutes/2007-4285m1-Final.pdf

Conflicts: Suellen M. Walker reported no conflicts of interest

Conflicts: Tony L Yaksh reported no conflicts of interest

Contributor Information

Suellen M. Walker, Portex Unit: Pain Research, UCL Institute of Child Health and Great Ormond St Hospital NHS Trust. Role: This author co-wrote the manuscript.

Tony L. Yaksh, Department of Anesthesiology, University of California San Diego, La Jolla, California, USA. Role: This author co-wrote the manuscript.

References

1. Anand KJ, Sippell WG, Aynsley-Green A. Randomised trial of fentanyl anaesthesia in preterm babies undergoing surgery: effects on the stress response. Lancet. 1987;1:62–6.
2. Anand KJ, Sippell WG, Schofield NM, Aynsley-Green A. Does halothane anaesthesia decrease the metabolic and endocrine stress responses of newborn infants undergoing operation? Br Med J (Clin Res Ed) 1988;296:668–72. [PMC free article]
3. Hermann C, Hohmeister J, Demirakca S, Zohsel K, Flor H. Long-term alteration of pain sensitivity in school-aged children with early pain experiences. Pain. 2006;125:278–85.
4. Hohmeister J, Demirakca S, Zohsel K, Flor H, Hermann C. Responses to pain in school-aged children with experience in a neonatal intensive care unit: cognitive aspects and maternal influences. Eur J Pain. 2009;13:94–101.
5. Walker SM, Franck LS, Fitzgerald M, Myles J, Stocks J, Marlow N. Long-term impact of neonatal intensive care and surgery on somatosensory perception in children born extremely preterm. Pain. 2009;141:79–87.
6. Schmelzle-Lubiecki BM, Campbell KA, Howard RH, Franck L, Fitzgerald M. Long-term consequences of early infant injury and trauma upon somatosensory processing. Eur J Pain. 2007;11:799–809.
7. Peters JW, Schouw R, Anand KJ, van Dijk M, Duivenvoorden HJ, Tibboel D. Does neonatal surgery lead to increased pain sensitivity in later childhood? Pain. 2005;114:444–54.
8. Grunau RE, Holsti L, Peters JW. Long-term consequences of pain in human neonates. Semin Fetal Neonatal Med. 2006;11:268–75.
9. Taddio A, Katz J. The effects of early pain experience in neonates on pain responses in infancy and childhood. Paediatr Drugs. 2005;7:245–57.
10. Jevtovic-Todorovic V, Hartman RE, Izumi Y, Benshoff ND, Dikranian K, Zorumski CF, Olney JW, Wozniak DF. Early exposure to common anesthetic agents causes widespread neurodegeneration in the developing rat brain and persistent learning deficits. J Neurosci. 2003;23:876–82.
11. Stratmann G. Review article: neurotoxicity of anesthetic drugs in the developing brain. Anesth Analg. 2011;113:1170–9.
12. Paule MG, Li M, Allen RR, Liu F, Zou X, Hotchkiss C, Hanig JP, Patterson TA, Slikker W, Jr, Wang C. Ketamine anesthesia during the first week of life can cause long-lasting cognitive deficits in rhesus monkeys. Neurotoxicol Teratol. 2011;33:220–30. [PMC free article]
13. Williams RK, Adams DC, Aladjem EV, Kreutz JM, Sartorelli KH, Vane DW, Abajian JC. The safety and efficacy of spinal anesthesia for surgery in infants: the Vermont Infant Spinal Registry. Anesth Analg. 2006;102:67–71.
14. Uguralp S, Mutus M, Koroglu A, Gurbuz N, Koltuksuz U, Demircan M. Regional anesthesia is a good alternative to general anesthesia in pediatric surgery: Experience in 1,554 children. J Pediatr Surg. 2002;37:610–3.
15. Tsui BC, Berde CB. Caudal analgesia and anesthesia techniques in children. Curr Opin Anaesthesiol. 2005;18:283–8.
16. Ivani G, Mossetti V. Continuous central and perineural infusions for postoperative pain control in children. Curr Opin Anaesthesiol. 2010;23:637–42.
17. Ecoffey C, Lacroix F, Giaufre E, Orliaguet G, Courreges P. Epidemiology and morbidity of regional anesthesia in children: a follow-up one-year prospective survey of the French-Language Society of Paediatric Anaesthesiologists (ADARPEF) Paediatr Anaesth. 2010;20:1061–9.

18. Rochette A, Dadure C, Raux O, Troncin R, Mailhee P, Capdevila X. A review of pediatric regional anesthesia practice during a 17-year period in a single institution. Paediatr Anaesth. 2007;17:874–80.
19. Kachko L, Simhi E, Tzeitlin E, Efrat R, Tarabikin E, Peled E, Metzner I, Katz J. Spinal anesthesia in neonates and infants - a single-center experience of 505 cases. Paediatr Anaesth. 2007;17:647–53.
20. Imbelloni LE, Vieira EM, Sperni F, Guizellini RH, Tolentino AP. Spinal anesthesia in children with isobaric local anesthetics: report on 307 patients under 13 years of age. Paediatr Anaesth. 2006;16:43–8.
21. Nickel US, Meyer RR, Brambrink AM. Spinal anesthesia in an extremely low birth weight infant. Paediatr Anaesth. 2005;15:58–62.
22. Humphreys N, Bays SM, Parry AJ, Pawade A, Heyderman RS, Wolf AR. Spinal anesthesia with an indwelling catheter reduces the stress response in pediatric open heart surgery. Anesthesiology. 2005;103:1113–20.
23. Williams RK, McBride WJ, Abajian JC. Combined spinal and epidural anaesthesia for major abdominal surgery in infants. Can J Anaesth. 1997;44:511–4.
24. Somri M, Tome R, Yanovski B, Asfandiarov E, Carmi N, Mogilner J, David B, Gaitini LA. Combined spinal-epidural anesthesia in major abdominal surgery in high-risk neonates and infants. Paediatr Anaesth. 2007;17:1059–65.
25. Hammer GB, Ramamoorthy C, Cao H, Williams GD, Boltz MG, Kamra K, Drover DR. Postoperative analgesia after spinal blockade in infants and children undergoing cardiac surgery. Anesth Analg. 2005;100:1283–8.
26. Rochette A, Raux O, Troncin R, Dadure C, Verdier R, Capdevila X. Clonidine prolongs spinal anesthesia in newborns: a prospective dose-ranging study. Anesth Analg. 2004;98:56–9.
27. Rochette A, Troncin R, Raux O, Dadure C, Lubrano JF, Barbotte E, Capdevila X. Clonidine added to bupivacaine in neonatal spinal anesthesia: a prospective comparison in 124 preterm and term infants. Paediatr Anaesth. 2005;15:1072–7.
28. Henderson K, Sethna NF, Berde CB. Continuous caudal anesthesia for inguinal hernia repair in former preterm infants. J Clin Anesth. 1993;5:129–33.
29. Tobias J, Rasmussen G, Holcomb Gr, Brock, Morgan Wr. Continuous caudal anaesthesia with chloroprocaine as an adjunct to general anaesthesia in neonates. Can J Anaesth. 1996;43:69–72.
30. Murrell D, Gibson PR, Cohen RC. Continuous epidural analgesia in newborn infants undergoing major surgery. J Pediatr Surg. 1993;28:548–52.
31. Golianu B, Hammer GB. Pain management for pediatric thoracic surgery. Curr Opin Anaesthesiol. 2005;18:13–21.
32. Peutrell JM, Lonnqvist PA. Neuraxial blocks for anaesthesia and analgesia in children. Current Opinion in Anaesthesiology. 2003;16:461–470.
33. Willschke H, Bosenberg A, Marhofer P, Willschke J, Schwindt J, Weintraud M, Kapral S, Kettner S. Epidural catheter placement in neonates: sonoanatomy and feasibility of ultrasonographic guidance in term and preterm neonates. Reg Anesth Pain Med. 2007;32:34–40.
34. Giaufre E, Dalens B, Gombert A. Epidemiology and morbidity of regional anesthesia in children: a one-year prospective survey of the French-Language Society of Pediatric Anesthesiologists. Anesth Analg. 1996;83:904–12.
35. Llewellyn N, Moriarty A. The national pediatric epidural audit. Paediatr Anaesth. 2007;17:520–33.
36. Craven PD, Badawi N, Henderson-Smart DJ, O'Brien M. Regional (spinal, epidural, caudal) versus general anaesthesia in preterm infants undergoing inguinal herniorrhaphy in early infancy. Cochrane Database Syst Rev. 2003:CD003669.

37. Kost-Byerly S, Jackson EV, Yaster M, Kozlowski LJ, Mathews RI, Gearhart JP. Perioperative anesthetic and analgesic management of newborn bladder exstrophy repair. J Pediatr Urol. 2008;4:280–5.
38. Raghavan M, Montgomerie J. Anaesthetic management of gastroschisis - a review of our practice over the past 5 years. Paediatr Anaesth. 2008;18:731–5.
39. Vila R, Marhuenda C, Goncalves A, Gil-Jaurena JM, Pellicer M, Suescum MC, Miro L. Epidural analgesia in the surgery of congenital tracheal stenosis: slide tracheoplasty on cardiopulmonary bypass. Paediatr Anaesth. 2006;16:693–6.
40. Williams RK, Abajian JC. High spinal anaesthesia for repair of patent ductus arteriosus in neonates. Paediatr Anaesth. 1997;7:205–9.
41. Chalkiadis G. The rise and fall of continuous epidural infusions in children. Paediatr Anaesth. 2003;13:91–3.
42. Taenzer AH, Clark C. Efficacy of postoperative epidural analgesia in adolescent scoliosis surgery: a meta-analysis. Paediatr Anaesth. 2010;20:135–43.
43. Brouwers M, Driessen J, Severijnen R. Clinical letter: epidural analgesia in a newborn with Hirschsprung's disease, associated with congenital central hypoventilation syndrome. Eur J Anaesthesiol. 2000;17:751–3.
44. Sartorelli KH, Abajian JC, Kreutz JM, Vane DW. Improved outcome utilizing spinal anesthesia in high-risk infants. J Pediatr Surg. 1992;27:1022–5.
45. Stuth EA, Berens RJ, Staudt SR, Robertson FA, Scott JP, Stucke AG, Hoffman GM, Troshynski TJ, Tweddell JS, Zuperku EJ. The effect of caudal vs intravenous morphine on early extubation and postoperative analgesic requirements for stage 2 and 3 single-ventricle palliation: a double blind randomized trial. Paediatr Anaesth. 2011;21:441–53.
46. Aspirot A, Puligandla PS, Bouchard S, Su W, Flageole H, Laberge JM. A contemporary evaluation of surgical outcome in neonates and infants undergoing lung resection. J Pediatr Surg. 2008;43:508–12.
47. McNeely J, Farber N, Rusy L, Hoffman G. Epidural analgesia improves outcome following pediatric fundoplication. A retrospective analysis. Reg Anesth. 1997;22:16–23.
48. Cass LJ, Howard RF. Respiratory complications due to inadequate analgesia following thoracotomy in a neonate. Anaesthesia. 1994;49:879–80.
49. Raghavendran S, Diwan R, Shah T, Vas L. Continuous caudal epidural analgesia for congenital lobar emphysema: a report of three cases. Anesth Analg. 2001;93:348–50. 3rd contents page.
50. Solak M, Ulusoy H, Sarihan H. Effects of caudal block on cortisol and prolactin responses to postoperative pain in children. Eur J Pediatr Surg. 2000;10:219–23.
51. Sendasgupta C, Makhija N, Kiran U, Choudhary SK, Lakshmy R, Das SN. Caudal epidural sufentanil and bupivacaine decreases stress response in paediatric cardiac surgery. Ann Card Anaesth. 2009;12:27–33.
52. Wolf AR, Eyres RL, Laussen PC, Edwards J, Stanley IJ, Rowe P, Simon L. Effect of extradural analgesia on stress responses to abdominal surgery in infants. Br J Anaesth. 1993;70:654–60.
53. Somri M, Gaitini LA, Vaida SJ, Malatzkey S, Sabo E, Yudashkin M, Tome R. The effectiveness and safety of spinal anaesthesia in the pyloromyotomy procedure. Paediatr Anaesth. 2003;13:32–7.
54. Katznelson R, Mishaly D, Hegesh T, Perel A, Keidan I. Spinal anesthesia for diagnostic cardiac catheterization in high-risk infants. Paediatr Anaesth. 2005;15:50–3.
55. Lin YC, Sentivany-Collins SK, Peterson KL, Boltz MG, Krane EJ. Outcomes after single injection caudal epidural versus continuous infusion epidural via caudal approach for postoperative analgesia in infants and children undergoing patent ductus arteriosus ligation. Paediatr Anaesth. 1999;9:139–43.
56. Flandin-Blety C, Barrier G. Accidents following extradural analgesia in children. The results of a retrospective study. Paediatr Anaesth. 1995;5:41–6.

57. van Niekerk J, Bax-Vermeire BM, Geurts JW, Kramer PP. Epidurography in premature infants. Anaesthesia. 1990;45:722–5.
58. Kost-Byerly S, Tobin JR, Greenberg RS, Billett C, Zahurak M, Yaster M. Bacterial colonization and infection rate of continuous epidural catheters in children. Anesth Analg. 1998;86:712–6.
59. Strafford MA, Wilder RT, Berde CB. The risk of infection from epidural analgesia in children: a review of 1620 cases. Anesth Analg. 1995;80:234–8.
60. Sethna NF, Clendenin D, Athiraman U, Solodiuk J, Rodriguez DP, Zurakowski D. Incidence of epidural catheter-associated infections after continuous epidural analgesia in children. Anesthesiology. 2010;113:224–32.
61. Breschan C, Krumpholz R, Jost R, Likar R. Intraspinal haematoma following lumbar epidural anaesthesia in a neonate. Paediatr Anaesth. 2001;11:105–8.
62. Ecoffey C, Samii K. Neurologic complication after epidural anesthesia in a 15-year-old boy. Ann Fr Anesth Reanim. 1990;9:398.
63. Allison CE, Aronson DC, Geukers VG, van den Berg R, Schlack WS, Hollmann MW. Paraplegia after thoracotomy under combined general and epidural anesthesia in a child. Paediatr Anaesth. 2008;18:539–42.
64. Yigit NA, Bagbanci B, Celebi H. Drop foot after pediatric urological surgery under general and epidural anesthesia. Anesth Analg. 2006;103:1616.
65. Zeidan A, Narchi P, Goujard E, Benhamou D. Postoperative nerve irritation syndrome after epidural analgesia in a six-year-old child. Br J Anaesth. 2004;92:146–8.
66. Thammasitboon S, Rosen DA, Lutfi R, Ely BA, Weber MA, Hilvers PN, Gustafson RA. An institutional experience with epidural analgesia in children and young adults undergoing cardiac surgery. Paediatr Anaesth. 2010;20:720–6.
67. Lacroix F. Epidemiology and morbidity of regional anaesthesia in children. Curr Opin Anaesthesiol. 2008;21:345–9.
68. Valois T, Otis A, Ranger M, Muir JG. Incidence of self-limiting back pain in children following caudal blockade: an exploratory study. Paediatr Anaesth. 2010;20:844–50.
69. Dalens BJ, Mazoit JX. Adverse effects of regional anaesthesia in children. Drug Saf. 1998;19:251–68.
70. Gunter J. Benefit and risks of local anesthetics in infants and children. Paediatr Drugs. 2002;4:649–72.
71. Berde CB. Convulsions associated with pediatric regional anesthesia. Anesth Analg. 1992;75:164–6.
72. Bosenberg AT, Cronje L, Thomas J, Lopez T, Crean PM, Gustafsson U, Huledal G, Larsson LE. Ropivacaine plasma levels and postoperative analgesia in neonates and infants during 48–72h continuous epidural infusion following major surgery. Paediatr Anaesth. 2003;13:851–852.
73. McCann M, Sethna N, Mazoit J, Sakamoto M, Rifai N, Hope T, Sullivan L, Auble S, Berde C. The pharmacokinetics of epidural ropivacaine in infants and young children. Anesth Analg. 2001;93:893–7.
74. Hansen TG, Ilett KF, Reid C, Lim SI, Hackett LP, Bergesio R. Caudal ropivacaine in infants: population pharmacokinetics and plasma concentrations. Anesthesiology. 2001;94:579–84.
75. Larsson B, Lonnqvist P, Olsson G. Plasma concentrations of bupivacaine in neonates after continuous epidural infusion. Anesth Analg. 1997;84:501–5.
76. Yahalom B, Athiraman U, Soriano SG, Zurakowski D, Carpino EA, Corfas G, Berde CB. Spinal anesthesia in infant rats: development of a model and assessment of neurologic outcomes. Anesthesiology. 2011;114:1325–35. [PMC free article]
77. Williams DG, Howard RF. Epidural analgesia in children. A survey of current opinions and practices amongst UK paediatric anaesthetists. Paediatr Anaesth. 2003;13:769–76.
78. Howard RF, Carter B, Curry J, Morton N, Rivett K, Rose M, Tyrrell J, Walker SM, Williams DG. Association of Paediatric Anaesthetists: Good Practice in Postoperative and Procedural Pain. Pediatric Anesthesia. 2008;18(Suppl 1):1–81.

79. Ansermino M, Basu R, Vandebeek C, Montgomery C. Nonopioid additives to local anaesthetics for caudal blockade in children: a systematic review. Paediatr Anaesth. 2003;13:561–73.
80. Walker SM. Pain in children: recent advances and ongoing challenges. Br J Anaesth. 2008;101:101–10.
81. Anand KJ, Aranda JV, Berde CB, Buckman S, Capparelli EV, Carlo WA, Hummel P, Lantos J, Johnston CC, Lehr VT, Lynn AM, Maxwell LG, Oberlander TF, Raju TN, Soriano SG, Taddio A, Walco GA. Analgesia and anesthesia for neonates: study design and ethical issues. Clin Ther. 2005;27:814–43.
82. Disma N, Frawley G, Mameli L, Pistorio A, Alberighi OD, Montobbio G, Tuo P. Effect of epidural clonidine on minimum local anesthetic concentration (ED50) of levobupivacaine for caudal block in children. Paediatr Anaesth. 2011;21:128–35.
83. Johnston P, Findlow D, Aldridge LM, Doyle E. The effect of ketamine on 0.25% and 0. 125% bupivacaine for caudal epidural blockade in children. Paediatr Anaesth. 1999;9:31–4.
84. Schnabel A, Poepping DM, Kranke P, Zahn PK, Pogatzki-Zahn EM. Efficacy and adverse effects of ketamine as an additive for paediatric caudal anaesthesia: a quantitative systematic review of randomized controlled trials. Br J Anaesth. 2011;107:601–11.
85. Schnabel A, Poepping DM, Pogatzki-Zahn EM, Zahn PK. Efficacy and safety of clonidine as additive for caudal regional anesthesia: a quantitative systematic review of randomized controlled trials. Paediatr Anaesth. 2011;21:1219–30.
86. Engelman E, Marsala C. Bayesian enhanced meta-analysis of post-operative analgesic efficacy of additives for caudal analgesia in children. Acta Anaesthesiol Scand. 2012 Epub Feb 7.
87. Dahmani S, Michelet D, Abback PS, Wood C, Brasher C, Nivoche Y, Mantz J. Ketamine for perioperative pain management in children: a meta-analysis of published studies. Paediatr Anaesth. 2011;21:636–52.
88. Wolf AR, Hughes D, Hobbs AJ, Prys-Roberts C. Combined morphine-bupivacaine caudals for reconstructive penile surgery in children: systemic absorption of morphine and postoperative analgesia. Anaesth Intensive Care. 1991;19:17–21.
89. Sharpe P, Klein JR, Thompson JP, Rushman SC, Sherwin J, Wandless JG, Fell D. Analgesia for circumcision in a paediatric population: comparison of caudal bupivacaine alone with bupivacaine plus two doses of clonidine. Paediatr Anaesth. 2001;11:695–700.
90. Singh R, Kumar N, Singh P. Randomized controlled trial comparing morphine or clonidine with bupivacaine for caudal analgesia in children undergoing upper abdominal surgery. Br J Anaesth. 2011;106:96–100.
91. Wolf AR, Hughes D, Wade A, Mather SJ, Prys-Roberts C. Postoperative analgesia after paediatric orchidopexy: evaluation of a bupivacaine-morphine mixture. Br J Anaesth. 1990;64:430–5.
92. Castillo-Zamora C, Castillo-Peralta LA, Nava-Ocampo AA. Dose minimization study of single-dose epidural morphine in patients undergoing hip surgery under regional anesthesia with bupivacaine. Paediatr Anaesth. 2005;15:29–36.
93. Apiliogullari S, Duman A, Gok F, Akillioglu I, Ciftci I. Efficacy of a low-dose spinal morphine with bupivacaine for postoperative analgesia in children undergoing hypospadias repair. Paediatr Anaesth. 2009;19:1078–83.
94. Gall O, Aubineau JV, Berniere J, Desjeux L, Murat I. Analgesic effect of low-dose intrathecal morphine after spinal fusion in children. Anesthesiology. 2001;94:447–52.
95. Ganesh A, Kim A, Casale P, Cucchiaro G. Low-dose intrathecal morphine for postoperative analgesia in children. Anesth Analg. 2007;104:271–6.
96. Cousins MJ, Mather LE. Intrathecal and epidural administration of opioids. Anesthesiology. 1984;61:276–310.

97. Locatelli BG, Frawley G, Spotti A, Ingelmo P, Kaplanian S, Rossi B, Monia L, Sonzogni V. Analgesic effectiveness of caudal levobupivacaine and ketamine. Br J Anaesth. 2008;100:701–6.
98. Hager H, Marhofer P, Sitzwohl C, Adler L, Kettner S, Semsroth M. Caudal clonidine prolongs analgesia from caudal S(+)-ketamine in children. Anesth Analg. 2002;94:1169–72.
99. Herd D, Anderson BJ. Ketamine disposition in children presenting for procedural sedation and analgesia in a children's emergency department. Paediatr Anaesth. 2007;17:622–9.
100. Lin C, Durieux ME. Ketamine and kids: an update. Paediatr Anaesth. 2005;15:91–7.
101. Lois F, De Kock M. Something new about ketamine for pediatric anesthesia? Curr Opin Anaesthesiol. 2008;21:340–4.
102. Koinig H, Marhofer P, Krenn CG, Klimscha W, Wildling E, Erlacher W, Nikolic A, Turnheim K, Semsroth M. Analgesic effects of caudal and intramuscular S(+)-ketamine in children. Anesthesiology. 2000;93:976–80.
103. Gunes Y, Gunduz M, Unlugenc H, Ozalevli M, Ozcengiz D. Comparison of caudal vs intravenous tramadol administered either preoperatively or postoperatively for pain relief in boys. Paediatr Anaesth. 2004;14:324–8.
104. Cao JP, Miao XY, Liu J, Shi XY. An evaluation of intrathecal bupivacaine combined with intrathecal or intravenous clonidine in children undergoing orthopedic surgery: a randomized double-blinded study. Paediatr Anaesth. 2011;21:399–405.
105. Akin A, Ocalan S, Esmaoglu A, Boyaci A. The effects of caudal or intravenous clonidine on postoperative analgesia produced by caudal levobupivacaine in children. Paediatr Anaesth. 2010;20:350–5.
106. Gauntlett I. A comparison between local anaesthetic dorsal nerve block and caudal bupivacaine with ketamine for paediatric circumcision. Paediatr Anaesth. 2003;13:38–42.
107. Margetts L, Carr A, McFadyen G, Lambert A. A comparison of caudal bupivacaine and ketamine with penile block for paediatric circumcision. Eur J Anaesthesiol. 2008;25:1009–13.
108. Walker SM, Goudas LC, Cousins MJ, Carr DB. Combination spinal analgesic chemotherapy: a systematic review. Anesth Analg. 2002;95:674–715.
109. Sanders JC. Paediatric regional anaesthesia, a survey of practice in the United Kingdom. Br J Anaesth. 2002;89:707–10.
110. Menzies R, Congreve K, Herodes V, Berg S, Mason DG. A survey of pediatric caudal extradural anesthesia practice. Paediatr Anaesth. 2009;19:829–36.
111. Eich C, Strauss J. Prompt and powerful effect of a practice guideline on caudal additives. Paediatr Anaesth. 2009;19:271–2.
112. Ozbek H, Bilen A, Ozcengiz D, Gunes Y, Ozalevli M, Akman H. The comparison of caudal ketamine, alfentanil and ketamine plus alfentanil administration for postoperative analgesia in children. Paediatr Anaesth. 2002;12:610–6.
113. Goodarzi M. The advantages of intrathecal opioids for spinal fusion in children. Paediatr Anaesth. 1998;8:131–4.
114. De Mey JC, Strobbet J, Poelaert J, Hoebeke P, Mortier E. The influence of sufentanil and/or clonidine on the duration of analgesia after a caudal block for hypospadias repair surgery in children. Eur J Anaesthesiol. 2000;17:379–82.
115. Erol A, Tavlan A, Tuncer S, Topal A, Yurtcu M, Reisli R, Otelcioglu S. Caudal anesthesia for minor subumbilical pediatric surgery: a comparison of levobupivacaine alone and levobupivacaine plus sufentanil. J Clin Anesth. 2008;20:442–6.
116. Khan FA, Memon GA, Kamal RS. Effect of route of buprenorphine on recovery and postoperative analgesic requirement in paediatric patients. Paediatr Anaesth. 2002;12:786–90.
117. Lawhorn C, Brown R. Epidural morphine with butorphanol in pediatric patients. J Clin Anesth. 1994;6:91–4.

118. Lawhorn C, Stoner J, Schmitz M, Brown RJ, Stewart F, Volpe P, Shirey R. Caudal epidural butorphanol plus bupivacaine versus bupivacaine in pediatric outpatient genitourinary procedures. J Clin Anesth. 1997;9:103–8.
119. Szabova A, Sadhasivam S, Wang Y, Nick TG, Goldschneider K. Comparison of postoperative analgesia with epidural butorphanol/bupivacaine versus fentanyl/bupivacaine following pediatric urological procedures. J Opioid Manag. 2010;6:401–7.
120. Kelleher A, Black A, Penman S, Howard R. Comparison of caudal bupivacaine and diamorphine with caudal bupivacaine alone for repair of hypospadias. Br J Anaesth. 1996;77:586–90.
121. Moriarty A. Postoperative extradural infusions in children: preliminary data from a comparison of bupivacaine/diamorphine with plain ropivacaine. Paediatr Anaesth. 1999;9:423–7.
122. Vetter TR, Carvallo D, Johnson JL, Mazurek MS, Presson RG., Jr A comparison of single-dose caudal clonidine, morphine, or hydromorphone combined with ropivacaine in pediatric patients undergoing ureteral reimplantation. Anesth Analg. 2007;104:1356–63.
123. Demiraran Y, Kocaman B, Akman R. A comparison of the postoperative analgesic efficacy of single-dose epidural tramadol versus morphine in children. Br J Anaesth. 2005;95:510–3.
124. Senel AC, Akyol A, Dohman D, Solak M. Caudal bupivacaine-tramadol combination for postoperative analgesia in pediatric herniorrhaphy. Acta Anaesthesiol Scand. 2001;45:786–9.
125. Ozcengiz D, Gunduz M, Ozbek H, Isik G. Comparison of caudal morphine and tramadol for postoperative pain control in children undergoing inguinal herniorrhaphy. Paediatr Anaesth. 2001;11:459–64.
126. Prosser DP, Davis A, Booker PD, Murray A. Caudal tramadol for postoperative analgesia in pediatric hypospadias surgery. Br J Anaesth. 1997;79:293–6.
127. Batra YK, Prasad MK, Arya VK, Chari P, Yaddanapudi LN. Comparison of caudal tramadol vs bupivacaine for post-operative analgesia in children undergoing hypospadias surgery. Int J Clin Pharmacol Ther. 1999;37:238–42.
128. Bouchut JC, Dubois R, Godard J. Clonidine in preterm-infant caudal anesthesia may be responsible for postoperative apnea. Reg Anesth Pain Med. 2001;26:83–5.
129. Breschan C, Krumpholz R, Likar R, Kraschl R, Schalk HV. Can a dose of 2microg.kg(−1) caudal clonidine cause respiratory depression in neonates? Paediatr Anaesth. 1999;9:81–3.
130. Fellmann C, Gerber AC, Weiss M. Apnoea in a former preterm infant after caudal bupivacaine with clonidine for inguinal herniorrhaphy. Paediatr Anaesth. 2002;12:637–40.
131. De Negri P, Ivani G, Visconti C, De Vivo P, Lonnqvist PA. The dose-response relationship for clonidine added to a postoperative continuous epidural infusion of ropivacaine in children. Anesth Analg. 2001;93:71–6.
132. Klamt JG, Garcia LV, Stocche RM, Meinberg AC. Epidural infusion of clonidine or clonidine plus ropivacaine for postoperative analgesia in children undergoing major abdominal surgery. J Clin Anesth. 2003;15:510–4.
133. Saadawy I, Boker A, Elshahawy MA, Almazrooa A, Melibary S, Abdellatif AA, Afifi W. Effect of dexmedetomidine on the characteristics of bupivacaine in a caudal block in pediatrics. Acta Anaesthesiol Scand. 2009;53:251–6.
134. El-Hennawy AM, Abd-Elwahab AM, Abd-Elmaksoud AM, El-Ozairy HS, Boulis SR. Addition of clonidine or dexmedetomidine to bupivacaine prolongs caudal analgesia in children. Br J Anaesth. 2009;103:268–74.
135. Konakci S, Adanir T, Yilmaz G, Rezanko T. The efficacy and neurotoxicity of dexmedetomidine administered via the epidural route. Eur J Anaesthesiol. 2008;25:403–9.
136. Walker SM, Yaksh TL. New caudal additives in children: benefit vs. risk? Acta Anaesthesiol Scand. 2009;53:1097–8.
137. Weber F, Wulf H. Caudal bupivacaine and s(+)-ketamine for postoperative analgesia in children. Paediatr Anaesth. 2003;13:244–8.

138. Semple D, Findlow D, Aldridge LM, Doyle E. The optimal dose of ketamine for caudal epidural blockade in children. Anaesthesia. 1996;51:1170–2.
139. Panjabi N, Prakash S, Gupta P, Gogia AR. Efficacy of three doses of ketamine with bupivacaine for caudal analgesia in pediatric inguinal herniotomy. Reg Anesth Pain Med. 2004;29:28–31.
140. Cook B, Grubb DJ, Aldridge LA, Doyle E. Comparison of the effects of adrenaline, clonidine and ketamine on the duration of caudal analgesia produced by bupivacaine in children. Br J Anaesth. 1995;75:698–701.
141. Passariello M, Almenrader N, Canneti A, Rubeo L, Haiberger R, Pietropaoli P. Caudal analgesia in children: S(+)-ketamine vs S(+)-ketamine plus clonidine. Paediatr Anaesth. 2004;14:851–5.
142. Martindale S, Dix P, Stoddart P. Double-blind randomized controlled trial of caudal versus intravenous S(+)-ketamine for supplementation of caudal analgesia in children. Br J Anaesth. 2004;92:344–7.
143. De Negri P, Ivani G, Visconti C, De Vivo P. How to prolong postoperative analgesia after caudal anaesthesia with ropivacaine in children: S-ketamine versus clonidine. Paediatr Anaesth. 2001;11:679–83.
144. Marhofer P, Krenn CG, Plochl W, Wallner T, Glaser C, Koinig H, Fleischmann E, Hochtl A, Semsroth M. S(+)-ketamine for caudal block in paediatric anaesthesia. Br J Anaesth. 2000;84:341–5.
145. Odes R, Erhan OL, Demirci M, Goksu H. Effects of ketamine added to ropivacaine in pediatric caudal block. Agri. 2010;22:53–60.
146. Gunduz M, Ozalevli M, Ozbek H, Ozcengiz D. Comparison of caudal ketamine with lidocaine or tramadol administration for postoperative analgesia of hypospadias surgery in children. Paediatr Anaesth. 2006;16:158–63.
147. Akbas M, Titiz TA, Ertugrul F, Akbas H, Melikoglu M. Comparison of the effect of ketamine added to bupivacaine and ropivacaine, on stress hormone levels and the duration of caudal analgesia. Acta Anaesthesiol Scand. 2005;49:1520–6.
148. Kumar P, Rudra A, Pan AK, Acharya A. Caudal additives in pediatrics: a comparison among midazolam, ketamine, and neostigmine coadministered with bupivacaine. Anesth Analg. 2005;101:69–73.
149. Findlow D, Aldridge L, Doyle E. Comparison of caudal block using bupivacaine and ketamine with ilioinguinal nerve block for orchidopexy in children. Anaesthesia. 1997;52:1110–3.
150. Eisenach JC, Yaksh TL. Epidural ketamine in healthy children--what's the point? Anesth Analg. 2003;96:626.
151. Cousins MJ, Miller RD. Intrathecal midazolam: an ethical editorial dilemma. Anesth Analg. 2004;98:1507–8.
152. Yaksh TL, Allen JW. The use of intrathecal midazolam in humans: a case study of process. Anesth Analg. 2004;98:1536–45.
153. Goresky GV. The clinical utility of epidural midazolam for inguinal hernia repair in children. Can J Anaesth. 1995;42:755–7.
154. Gulec S, Buyukkidan B, Oral N, Ozcan N, Tanriverdi B. Comparison of caudal bupivacaine, bupivacaine-morphine and bupivacaine-midazolam mixtures for postoperative analgesia in children. Eur J Anaesthesiol. 1998;15:161–5.
155. Naguib M, el Gammal M, Elhattab Y, Seraj M. Midazolam for caudal analgesia in children: comparison with caudal bupivacaine. Can J Anaesth. 1995;42:758–64.
156. Baris S, Karakaya D, Kelsaka E, Guldogus F, Ariturk E, Tur A. Comparison of fentanyl-bupivacaine or midazolam-bupivacaine mixtures with plain bupivacaine for caudal anaesthesia in children. Paediatr Anaesth. 2003;13:126–31.

157. Hong JY, Lee IH, Shin SK, Park EY, Ban SY, Cho JE, Kil HK. Caudal midazolam does not affect sevoflurane requirements and recovery in pediatric day-case hernioplasty. Acta Anaesthesiol Scand. 2008;52:1411–4.
158. Mahajan R, Batra YK, Grover VK, Kajal J. A comparative study of caudal bupivacaine and midazolam-bupivacaine mixture for post-operative analgesia in children undergoing genitourinary surgery. Int J Clin Pharmacol Ther. 2001;39:116–20.
159. Hood DD, Eisenach JC, Tuttle R. Phase I safety assessment of intrathecal neostigmine methylsulfate in humans. Anesthesiology. 1995;82:331–43.
160. Eisenach JC, Hood DD, Curry R. Phase I human safety assessment of intrathecal neostigmine containing methyl- and propylparabens. Anesth Analg. 1997;85:842–6.
161. Lonnqvist PA. Adjuncts to caudal block in children--Quo vadis? Br J Anaesth. 2005;95:431–3.
162. Karaaslan K, Gulcu N, Ozturk H, Sarpkaya A, Colak C, Kocoglu H. Two different doses of caudal neostigmine co-administered with levobupivacaine produces analgesia in children. Paediatr Anaesth. 2009;19:487–493.
163. Memis D, Turan A, Karamanlioglu B, Kaya G, Sut N, Pamukcu Z. Caudal neostigmine for postoperative analgesia in paediatric surgery. Paediatr Anaesth. 2003;13:324–8.
164. Mahajan R, Grover VK, Chari P. Caudal neostigmine with bupivacaine produces a dose-independent analgesic effect in children. Can J Anaesth. 2004;51:702–6.
165. Turan A, Memis D, Basaran UN, Karamanlioglu B, Sut N. Caudal ropivacaine and neostigmine in pediatric surgery. Anesthesiology. 2003;98:719–22.
166. Bhardwaj N, Yaddanapudi S, Ghai B, Wig J. Neostigmine does not prolong the duration of analgesia produced by caudal bupivacaine in children undergoing urethroplasty. J Postgrad Med. 2007;53:161–5.
167. Taheri R, Shayeghi S, Razavi SS, Sadeghi A, Ghabili K, Ghojazadeh M, Rouzrokh M. Efficacy of bupivacaine-neostigmine and bupivacaine-tramadol in caudal block in pediatric inguinal herniorrhaphy. Paediatr Anaesth. 2010;20:866–72.
168. Batra YK, Arya VK, Mahajan R, Chari P. Dose response study of caudal neostigmine for postoperative analgesia in paediatric patients undergoing genitourinary surgery. Paediatr Anaesth. 2003;13:515–21.
169. Abdulatif M, El-Sanabary M. Caudal neostigmine, bupivacaine, and their combination for postoperative pain management after hypospadias surgery in children. Anesth Analg. 2002;95:1215–8.
170. Almenrader N, Passariello M, D'Amico G, Haiberger R, Pietropaoli P. Caudal additives for postoperative pain management in children: S(+)-ketamine and neostigmine. Paediatr Anaesth. 2005;15:143–7.
171. Batra YK, Rajeev S, Panda NB, Lokesh VC, Rao KL. Intrathecal neostigmine with bupivacaine for infants undergoing lower abdominal and urogenital procedures: dose response. Acta Anaesthesiol Scand. 2009;53:470–5.
172. Eisenach JC, James FM, 3rd, Gordh T, Jr, Yaksh TL. New epidural drugs: primum non nocere. Anesth Analg. 1998;87:1211–2.
173. Eisenach JC, Yaksh TL. Safety in numbers: how do we study toxicity of spinal analgesics? Anesthesiology. 2002;97:1047–9.
174. Shafer SL. Anesthesia & Analgesia's Policy on Off-Label Drug Administration in Clinical Trials. Anesth Analg. 2007;105:13–15.
175. Rowbotham MC. Pain's policy on the spinal administration of drugs. Pain. 2010;149:415–6.
176. Eisenach JC, Shafer SL, Yaksh T. The need for a journal policy on intrathecal, epidural, and perineural administration of non-approved drugs. Pain. 2010;149:417–9.
177. Neal JM, Rathmell JP, Rowlingson JC. Publishing studies that involve "off-label" use of drugs: formalizing Regional Anesthesia and Pain Medicine's policy. Regional anesthesia and pain medicine. 2009;34:391–2.

178. Dalens B. Some current controversies in paediatric regional anaesthesia. Curr Opin Anaesthesiol. 2006;19:301–8.
179. de Beer DA, Thomas ML. Caudal additives in children--solutions or problems? Br J Anaesth. 2003;90:487–98.
180. Symons JA, Palmer GM. Neuropathic pain and foot drop related to nerve injury after short duration surgery and caudal analgesia. Clin J Pain. 2008;24:647–9.
181. Berde C. Regional anesthesia in children: what have we learned? Anesth Analg. 1996;83:897–900.
182. Taniguchi M, Bollen AW, Drasner K. Sodium bisulfite: scapegoat for chloroprocaine neurotoxicity? Anesthesiology. 2004;100:85–91.
183. Loo CC, Irestedt L. Cauda equina syndrome after spinal anaesthesia with hyperbaric 5% lignocaine: a review of six cases of cauda equina syndrome reported to the Swedish Pharmaceutical Insurance 1993–1997. Acta Anaesthesiol Scand. 1999;43:371–9.
184. Hylden JL, Wilcox GL. Intrathecal morphine in mice: a new technique. Eur J Pharmacol. 1980;67:313–6.
185. Stokes JA, Corr M, Yaksh TL. Transient tactile allodynia following intrathecal puncture in mouse: contributions of Toll-like receptor signaling. Neuroscience letters. 2011;504:215–8. [PMC free article]
186. Westin BD, Walker SM, Deumens R, Grafe M, Yaksh TL. Validation of a Preclinical Spinal Safety Model: Effects of Intrathecal Morphine in the Neonatal Rat. Anesthesiology. 2010;113:183–199. [PMC free article]
187. Hughes HE, Barr GA. Analgesic effects of intrathecally applied noradrenergic compounds in the developing rat: differences due to thermal vs mechanical nociception. Brain Res. 1988;469:109–20.
188. Howard RF, Hatch DJ, Cole TJ, Fitzgerald M. Inflammatory pain and hypersensitivity are selectively reversed by epidural bupivacaine and are developmentally regulated. Anesthesiology. 2001;95:421–7.
189. Marsh D, Dickenson A, Hatch D, Fitzgerald M. Epidural opioid analgesia in infant rats I: mechanical and heat responses. Pain. 1999;82:23–32.
190. Walker SM, Howard RF, Keay KA, Fitzgerald M. Developmental age influences the effect of epidural dexmedetomidine on inflammatory hyperalgesia in rat pups. Anesthesiology. 2005;102:1226–1234.
191. Johnson RA, Lopez MJ, Hendrickson DA, Kruse-Elliott KT. Cephalad distribution of three differing volumes of new methylene blue injected into the epidural space in adult goats. Vet Surg. 1996;25:448–51.
192. Lee I, Yamagishi N, Oboshi K, Yamada H. Distribution of new methylene blue injected into the lumbosacral epidural space in cats. Vet Anaesth Analg. 2004;31:190–4.
193. Lopez MJ, Johnson R, Hendrickson DA, Kruse-Elliott KT. Craniad migration of differing doses of new methylene blue injected into the epidural space after death of calves and juvenile pigs. Am J Vet Res. 1997;58:786–90.
194. Hathway G, Harrop E, Baccei M, Walker S, Moss A, Fitzgerald M. A postnatal switch in GABAergic control of spinal cutaneous reflexes. Eur J Neurosci. 2006;23:112–8. [PMC free article]
195. Allen JW, Horais KA, Tozier NA, Yaksh TL. Opiate pharmacology of intrathecal granulomas. Anesthesiology. 2006;105:590–8.
196. Walker SM, Westin BD, Deumens R, Grafe M, Yaksh TL. Effects of Intrathecal Ketamine in the Neonatal Rat: Evaluation of Apoptosis and Long-term Functional Outcome. Anesthesiology. 2010;113:147–159. [PMC free article]
197. Fitzgerald M, Walker SM. Infant pain management: a developmental neurobiological approach. Nat Clin Pract Neurol. 2009;5:35–50.

198. King TE, Barr GA. Spinal cord ionotropic glutamate receptors function in formalin-induced nociception in preweaning rats. Psychopharmacology (Berl) 2007;192:489–98.
199. Walker SM, Fitzgerald M. Characterization of spinal alpha-adrenergic modulation of nociceptive transmission and hyperalgesia throughout postnatal development in rats. Br J Pharmacol. 2007;151:1334–42. [PMC free article]
200. Nandi R, Beacham D, Middleton J, Koltzenburg M, Howard RF, Fitzgerald M. The functional expression of mu opioid receptors on sensory neurons is developmentally regulated; morphine analgesia is less selective in the neonate. Pain. 2004;111:38–50.
201. Marsh D, Dickenson A, Hatch D, Fitzgerald M. Epidural opioid analgesia in infant rats II: responses to carrageenan and capsaicin. Pain. 1999;82:33–8.
202. Barr GA, Miya DY, Paredes W. Analgesic effects of intraventricular and intrathecal injection of morphine and ketocyclazocine in the infant rat. Brain Res. 1992;584:83–91.
203. Rahman W, Dashwood MR, Fitzgerald M, Aynsley-Green A, Dickenson AH. Postnatal development of multiple opioid receptors in the spinal cord and development of spinal morphine analgesia. Brain Res Dev Brain Res. 1998;108:239–54.
204. Nandi R, Fitzgerald M. Opioid analgesia in the newborn. Eur J Pain. 2005;9:105–8.
205. Walker SM, Grafe M, Yaksh TL. Intrathecal Clonidine in the Neonatal Rat: Dose-Dependent Analgesia and Evaluation of Spinal Apoptosis and Toxicity. Anesth Analg. 2012 Epub Mar 30. [PMC free article]
206. Gehling M, Tryba M. Risks and side-effects of intrathecal morphine combined with spinal anaesthesia: a meta-analysis. Anaesthesia. 2009;64:643–51.
207. Rauck RL, Wallace MS, Leong MS, Minehart M, Webster LR, Charapata SG, Abraham JE, Buffington DE, Ellis D, Kartzinel R. A randomized, double-blind, placebo-controlled study of intrathecal ziconotide in adults with severe chronic pain. J Pain Symptom Manage. 2006;31:393–406.
208. Lu JK, Manullang TR, Staples MH, Kem SE, Balley PL. Maternal respiratory arrests, severe hypotension, and fetal distress after administration of intrathecal, sufentanil, and bupivacaine after intravenous fentanyl. Anesthesiology. 1997;87:170–2.
209. Yaksh TL, Horais KA, Tozier NA, Allen JW, Rathbun M, Rossi SS, Sommer C, Meschter C, Richter PJ, Hildebrand KR. Chronically infused intrathecal morphine in dogs. Anesthesiology. 2003;99:174–87.
210. Yaksh TL, Rathbun ML, Provencher JC. Preclinical safety evaluation for spinal drugs. In: Yaksh TL, editor. Spinal Drug Delivery. Amsterdam: Elsevier Science B.V; 1999. pp. 417–437.
211. Hensch TK. Critical period regulation. Annu Rev Neurosci. 2004;27:549–79.
212. Olney JW, Young C, Wozniak DF, Ikonomidou C, Jevtovic-Todorovic V. Anesthesia-induced developmental neuroapoptosis. Does it happen in humans? Anesthesiology. 2004;101:273–5.
213. Mellon RD, Simone AF, Rappaport BA. Use of anesthetic agents in neonates and young children. Anesth Analg. 2007;104:509–20.
214. Loepke AW, Soriano SG. An assessment of the effects of general anesthetics on developing brain structure and neurocognitive function. Anesth Analg. 2008;106:1681–707.
215. Davidson AJ. Anesthesia and neurotoxicity to the developing brain: the clinical relevance. Paediatr Anaesth. 2011;21:716–21.
216. Creeley CE, Olney JW. The young: neuroapoptosis induced by anesthetics and what to do about it. Anesth Analg. 2010;110:442–8.
217. Yon JH, Daniel-Johnson J, Carter LB, Jevtovic-Todorovic V. Anesthesia induces neuronal cell death in the developing rat brain via the intrinsic and extrinsic apoptotic pathways. Neuroscience. 2005;135:815–27.
218. Sanders RD, Xu J, Shu Y, Fidalgo A, Ma D, Maze M. General anesthetics induce apoptotic neurodegeneration in the neonatal rat spinal cord. Anesth Analg. 2008;106:1708–11.

219. Jackman A, Fitzgerald M. Development of peripheral hindlimb and central spinal cord innervation by subpopulations of dorsal root ganglion cells in the embryonic rat. J Comp Neurol. 2000;418:281–98.
220. Fitzgerald M, Butcher T, Shortland P. Developmental changes in the laminar termination of A fibre cutaneous sensory afferents in the rat spinal cord dorsal horn. J Comp Neurol. 1994;348:225–33.
221. Torsney C, Meredith-Middleton J, Fitzgerald M. Neonatal capsaicin treatment prevents the normal postnatal withdrawal of A fibres from lamina II without affecting fos responses to innocuous peripheral stimulation. Brain Res Dev Brain Res. 2000;121:55–65.
222. Beggs S, Torsney C, Drew LJ, Fitzgerald M. The postnatal reorganization of primary afferent input and dorsal horn cell receptive fields in the rat spinal cord is an activity-dependent process. Eur J Neurosci. 2002;16:1249–58.
223. Granmo M, Petersson P, Schouenborg J. Action-based body maps in the spinal cord emerge from a transitory floating organization. J Neurosci. 2008;28:5494–503.
224. Shortland P, Fitzgerald M. Neonatal sciatic nerve section results in a rearrangement of the central terminals of saphenous and axotomized sciatic nerve afferents in the dorsal horn of the spinal cord of the adult rat. Eur J Neurosci. 1994;6:75–86.
225. Coggeshall RE, Pover CM, Fitzgerald M. Dorsal root ganglion cell death and surviving cell numbers in relation to the development of sensory innervation in the rat hindlimb. Brain Res Dev Brain Res. 1994;82:193–212.
226. Himes BT, Tessler A. Death of some dorsal root ganglion neurons and plasticity of others following sciatic nerve section in adult and neonatal rats. J Comp Neurol. 1989;284:215–30.
227. Ren K, Anseloni V, Zou SP, Wade EB, Novikova SI, Ennis M, Traub RJ, Gold MS, Dubner R, Lidow MS. Characterization of basal and re-inflammation-associated long-term alteration in pain responsivity following short-lasting neonatal local inflammatory insult. Pain. 2004;110:588–96.
228. Chu YC, Chan KH, Tsou MY, Lin SM, Hsieh YC, Tao YX. Mechanical pain hypersensitivity after incisional surgery is enhanced in rats subjected to neonatal peripheral inflammation: effects of N-methyl-D-aspartate receptor antagonists. Anesthesiology. 2007;106:1204–12.
229. Walker SM, Tochiki KK, Fitzgerald M. Hindpaw incision in early life increases the hyperalgesic response to repeat surgical injury: critical period and dependence on initial afferent activity. Pain. 2009;147:99–106.
230. Beggs S, Currie G, Salter MW, Fitzgerald M, Walker SM. Priming of adult pain responses by neonatal pain experience: maintenance by central neuroimmune activity. Brain. 2012;135:404–17. [PMC free article]
231. Pattinson D, Fitzgerald M. The neurobiology of infant pain: development of excitatory and inhibitory neurotransmission in the spinal dorsal horn. Reg Anesth Pain Med. 2004;29:36–44.
232. Fitzgerald M. The development of nociceptive circuits. Nat Rev Neurosci. 2005;6:507–20.
233. Baccei ML. Development of pain: maturation of spinal inhibitory networks. Int Anesthesiol Clin. 2007;45:1–11.
234. Baccei ML, Fitzgerald M. Development of GABAergic and glycinergic transmission in the neonatal rat dorsal horn. J Neurosci. 2004;24:4749–57.
235. Bremner L, Fitzgerald M, Baccei M. Functional GABA(A)-receptor-mediated inhibition in the neonatal dorsal horn. J Neurophysiol. 2006;95:3893–7.
236. Bremner L, Fitzgerald M. Postnatal tuning of cutaneous inhibitory receptive fields in the rat. J Physiol. 2007 [PMC free article]
237. Koch SC, Fitzgerald M, Hathway GJ. Midazolam Potentiates Nociceptive Behavior, Sensitizes Cutaneous Reflexes, and Is Devoid of Sedative Action in Neonatal Rats. Anesthesiology. 2008;108:122–129.

238. Hathway GJ, Koch S, Low L, Fitzgerald M. The changing balance of brainstem-spinal cord modulation of pain processing over the first weeks of rat postnatal life. J Physiol. 2009;587:2927–35. [PMC free article]
239. Vutskits L, Gascon E, Tassonyi E, Kiss JZ. Effect of ketamine on dendritic arbor development and survival of immature GABAergic neurons in vitro. Toxicol Sci. 2006;91:540–9.
240. Vutskits L, Gascon E, Tassonyi E, Kiss JZ. Clinically relevant concentrations of propofol but not midazolam alter in vitro dendritic development of isolated gamma-aminobutyric acid-positive interneurons. Anesthesiology. 2005;102:970–6.
241. Wood SL, Beyer BK, Cappon GD. Species comparison of postnatal CNS development: functional measures. Birth defects research Part B, Developmental and reproductive toxicology. 2003;68:391–407.
242. Clancy B, Kersh B, Hyde J, Darlington RB, Anand KJ, Finlay BL. Web-based method for translating neurodevelopment from laboratory species to humans. Neuroinformatics. 2007;5:79–94.
243. Nagarajan R, Darlington RB, Finlay BL, Clancy B. ttime: an R package for translating the timing of brain development across mammalian species. Neuroinformatics. 2010;8:201–5. [PMC free article]
244. Clancy B, Finlay BL, Darlington RB, Anand KJ. Extrapolating brain development from experimental species to humans. Neurotoxicology. 2007;28:931–7. [PMC free article]
245. McCutcheon JE, Marinelli M. Age matters. Eur J Neurosci. 2009;29:997–1014. [PMC free article]
246. Brambrink AM, Evers AS, Avidan MS, Farber NB, Smith DJ, Zhang X, Dissen GA, Creeley CE, Olney JW. Isoflurane-induced neuroapoptosis in the neonatal rhesus macaque brain. Anesthesiology. 2010;112:834–41. [PMC free article]
247. Lauder GR, White MC. Neuropathic pain following multilevel surgery in children with cerebral palsy: a case series and review. Paediatr Anaesth. 2005;15:412–20.
248. Fitzgerald M, Shaw A, MacIntosh N. Postnatal development of the cutaneous flexor reflex: comparative study of preterm infants and newborn rat pups. Dev Med Child Neurol. 1988;30:520–6.
249. Andrews K, Fitzgerald M. Flexion reflex responses in biceps femoris and tibialis anterior in human neonates. Early Hum Dev. 2000;57:105–10.
250. Andrews KA, Desai D, Dhillon HK, Wilcox DT, Fitzgerald M. Abdominal sensitivity in the first year of life: comparison of infants with and without prenatally diagnosed unilateral hydronephrosis. Pain. 2002;100:35–46.
251. Andrews K, Fitzgerald M. The cutaneous withdrawal reflex in human neonates: sensitization, receptive fields, and the effects of contralateral stimulation. Pain. 1994;56:95–101.
252. Holmberg H, Schouenborg J. Postnatal development of the nociceptive withdrawal reflexes in the rat: a behavioural and electromyographic study. J Physiol. 1996;493(Pt 1):239–52. [PMC free article]
253. Schouenborg J. Modular organisation and spinal somatosensory imprinting. Brain Res Brain Res Rev. 2002;40:80–91.
254. Andrews K, Fitzgerald M. Cutaneous flexion reflex in human neonates: a quantitative study of threshold and stimulus-response characteristics after single and repeated stimuli. Dev Med Child Neurol. 1999;41:696–703.
255. Slater R, Cornelissen L, Fabrizi L, Patten D, Yoxen J, Worley A, Boyd S, Meek J, Fitzgerald M. Oral sucrose as an analgesic drug for procedural pain in newborn infants: a randomised controlled trial. Lancet. 2010;376:1225–32. [PMC free article]
256. Ikonomidou C, Bittigau P, Ishimaru MJ, Wozniak DF, Koch C, Genz K, Price MT, Stefovska V, Horster F, Tenkova T, Dikranian K, Olney JW. Ethanol-induced apoptotic neurodegeneration and fetal alcohol syndrome. Science. 2000;287:1056–60.

257. Lawson SJ, Davies HJ, Bennett JP, Lowrie MB. Evidence that spinal interneurons undergo programmed cell death postnatally in the rat. Eur J Neurosci. 1997;9:794–9.
258. Lowrie MB, Lawson SJ. Cell death of spinal interneurones. Prog Neurobiol. 2000;61:543–55.
259. de Louw AJ, de Vente J, Steinbusch HP, Gavilanes AW, Steinbusch HW, Blanco CE, Troost J, Vles JS. Apoptosis in the rat spinal cord during postnatal development; the effect of perinatal asphyxia on programmed cell death. Neuroscience. 2002;112:751–8.
260. Drasner K. Anesthetic Effects on the Developing Nervous System: If You Aren't Concerned, You Haven't Been Paying Attention. Anesthesiology. 2010;113:10–2.
261. Hashimoto K, Sakura S, Bollen AW, Ciriales R, Drasner K. Comparative toxicity of glucose and lidocaine administered intrathecally in the rat. Reg Anesth Pain Med. 1998;23:444–50.
262. Blaylock M, Engelhardt T, Bissonnette B. Fundamentals of neuronal apoptosis relevant to pediatric anesthesia. Paediatr Anaesth. 2010;20:383–95.
263. Jevtovic-Todorovic V, Olney JW. PRO: Anesthesia-induced developmental neuroapoptosis: status of the evidence. Anesth Analg. 2008;106:1659–63.
264. Schmued LC, Stowers CC, Scallet AC, Xu L. Fluoro-Jade C results in ultra high resolution and contrast labeling of degenerating neurons. Brain Res. 2005;1035:24–31.
265. Hampl KF, Schneider MC, Drasner K. Toxicity of spinal local anaesthetics. Curr Opin Anaesthesiol. 1999;12:559–64.
266. Kishimoto T, Bollen AW, Drasner K. Comparative spinal neurotoxicity of prilocaine and lidocaine. Anesthesiology. 2002;97:1250–3.
267. De Roo M, Klauser P, Briner A, Nikonenko I, Mendez P, Dayer A, Kiss JZ, Muller D, Vutskits L. Anesthetics rapidly promote synaptogenesis during a critical period of brain development. PLoS One. 2009;4:e7043. [PMC free article]
268. Vutskits L, Gascon E, Potter G, Tassonyi E, Kiss JZ. Low concentrations of ketamine initiate dendritic atrophy of differentiated GABAergic neurons in culture. Toxicology. 2007;234:216–26.
269. Melemedjian OK, Price TJ. Dendritic spine plasticity as an underlying mechanism of neuropathic pain: commentary on Tan et al. Exp Neurol. 2012;233:740–4. [PMC free article]
270. Tan AM, Waxman SG. Spinal cord injury, dendritic spine remodeling, and spinal memory mechanisms. Exp Neurol. 2012;235:142–51.
271. Pearn ML, Hu Y, Niesman IR, Patel HH, Drummond JC, Roth DM, Akassoglou K, Patel PM, Head BP. Propofol neurotoxicity is mediated by p75 neurotrophin receptor activation. Anesthesiology. 2012;116:352–61. [PMC free article]
272. Lemkuil BP, Head BP, Pearn ML, Patel HH, Drummond JC, Patel PM. Isoflurane neurotoxicity is mediated by p75NTR-RhoA activation and actin depolymerization. Anesthesiology. 2011;114:49–57. [PMC free article]
273. Morgan PG, Sedensky M. A new phase in anesthetic-induced neurotoxicity research. Anesthesiology. 2011;114:10–1.
274. Gold MS, Reichling DB, Hampl KF, Drasner K, Levine JD. Lidocaine toxicity in primary afferent neurons from the rat. J Pharmacol Exp Ther. 1998;285:413–21.
275. Lirk P, Haller I, Colvin HP, Frauscher S, Kirchmair L, Gerner P, Klimaschewski L. In vitro, lidocaine-induced axonal injury is prevented by peripheral inhibition of the p38 mitogen-activated protein kinase, but not by inhibiting caspase activity. Anesth Analg. 2007;105:1657–64.
276. Werdehausen R, Braun S, Hermanns H, Kremer D, Kury P, Hollmann MW, Bauer I, Stevens MF. The influence of adjuvants used in regional anesthesia on lidocaine-induced neurotoxicity in vitro. Reg Anesth Pain Med. 2011;36:436–43.
277. Williams BA, Hough KA, Tsui BY, Ibinson JW, Gold MS, Gebhart GF. Neurotoxicity of adjuvants used in perineural anesthesia and analgesia in comparison with ropivacaine. Reg Anesth Pain Med. 2011;36:225–30. [PMC free article]

278. Candido KD, Knezevic NN. All adjuvants to local anesthetics were not created equal: animal data evaluating neurotoxicity, thermal hyperalgesia, and relevance to human application. Reg Anesth Pain Med. 2011;36:211–2.
279. Hahn KA. Chemotherapy Dose Calculation and Administration in Exotic Animal Species. Seminars in Avian and Exotic Pet Medicine. 2005;14:193–198.
280. Reagan-Shaw S, Nihal M, Ahmad N. Dose translation from animal to human studies revisited. Faseb J. 2008;22:659–61.
281. U.S. Department of Health and Human Services Food and Drug Administration. Guidance for Industry Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers. Edited by Center for Drug Evaluation and Research (CDER). Office of Training and Communications Division of Drug Information, HFD-240. Center for Drug Evaluation and Research Food and Drug Administration; Rockville, MD: 2005.
282. Carpenter RL, Hogan QH, Liu SS, Crane B, Moore J. Lumbosacral cerebrospinal fluid volume is the primary determinant of sensory block extent and duration during spinal anesthesia. Anesthesiology. 1998;89:24–9.
283. Rigler ML, Drasner K, Krejcie TC, Yelich SJ, Scholnick FT, DeFontes J, Bohner D. Cauda equina syndrome after continuous spinal anesthesia. Anesth Analg. 1991;72:275–81.
284. McMillan MR, Doud T, Nugent W. Catheter-associated masses in patients receiving intrathecal analgesic therapy. Anesth Analg. 2003;96:186–90.
285. Hoederath P, Gautschi OP, Land M, Hildebrandt G, Fournier JY. Formation of two consecutive intrathecal catheter tip granulomas within nine months. Cen Eur Neurosurg. 2010;71:39–42.
286. Schnabel A, Zahn PK, Pogatzki-Zahn EM. Use of ketamine in children - what are the next steps? Paediatr Anaesth. 2011;21:1080–1.
287. Davidson AJ. Neurotoxicity and the need for anesthesia in the newborn: does the emperor have no clothes? Anesthesiology. 2012;116:507–9.
288. Koroglu A, Durmus M, Togal T, Ozpolat Z, Ersoy MO. Spinal anaesthesia in full-term infants of 0–6 months: are there any differences regarding age? Eur J Anaesthesiol. 2005;22:111–6.
289. Hermanns H, Stevens MF, Werdehausen R, Braun S, Lipfert P, Jetzek-Zader M. Sedation during spinal anaesthesia in infants. Br J Anaesth. 2006;97:380–4.
290. Kokki H, Tuovinen K, Hendolin H. Spinal anaesthesia for paediatric day-case surgery: a double-blind, randomized, parallel group, prospective comparison of isobaric and hyperbaric bupivacaine. Br J Anaesth. 1998;81:502–6.
291. William JM, Stoddart PA, Williams SA, Wolf AR. Post-operative recovery after inguinal herniotomy in ex-premature infants: comparison between sevoflurane and spinal anaesthesia. Br J Anaesth. 2001;86:366–71.
292. Frawley G, Skinner A, Thomas J, Smith S. Ropivacaine spinal anesthesia in neonates: a dose range finding study. Paediatr Anaesth. 2007;17:126–32.
293. Bosenberg AT, Thomas J, Cronje L, Lopez T, Crean PM, Gustafsson U, Huledal G, Larsson LE. Pharmacokinetics and efficacy of ropivacaine for continuous epidural infusion in neonates and infants. Paediatr Anaesth. 2005;15:739–49.
294. Wulf H, Peters C, Behnke H. The pharmacokinetics of caudal ropivacaine 0.2% in children. A study of infants aged less than 1 year and toddlers aged 1–5 years undergoing inguinal hernia repair. Anaesthesia. 2000;55:757–60.
295. Frawley GP, Farrell T, Smith S. Levobupivacaine spinal anesthesia in neonates: a dose range finding study. Paediatr Anaesth. 2004;14:838–44.
296. Chalkiadis GA, Anderson BJ, Tay M, Bjorksten A, Kelly JJ. Pharmacokinetics of levobupivacaine after caudal epidural administration in infants less than 3 months of age. Br J Anaesth. 2005;95:524–9.

297. Rice LJ, DeMars PD, Whalen TV, Crooms JC, Parkinson SK. Duration of spinal anesthesia in infants less than one year of age. Comparison of three hyperbaric techniques. Reg Anesth. 1994;19:325–9.
298. Viscomi CM, Abajian JC, Wald SL, Rathmell JP, Wilson JT. Spinal anesthesia for repair of meningomyelocele in neonates. Anesth Analg. 1995;81:492–5.
299. Shenkman Z, Hoppenstein D, Litmanowitz I, Shorer S, Gutermacher M, Lazar L, Erez I, Jedeikin R, Freud E. Spinal anesthesia in 62 premature, former-premature or young infants--technical aspects and pitfalls. Can J Anaesth. 2002;49:262–9.
300. Peterson KL, DeCampli WM, Pike NA, Robbins RC, Reitz BA. A report of two hundred twenty cases of regional anesthesia in pediatric cardiac surgery. Anesth Analg. 2000;90:1014–9.
301. Lejus C, Surbled M, Schwoerer D, Renaudin M, Guillaud C, Berard L, Pinaud M. Postoperative epidural analgesia with bupivacaine and fentanyl: hourly pain assessment in 348 paediatric cases. Paediatr Anaesth. 2001;11:327–32.
302. Batra YK, Lokesh VC, Panda NB, Rajeev S, Rao KL. Dose-response study of intrathecal fentanyl added to bupivacaine in infants undergoing lower abdominal and urologic surgery. Paediatr Anaesth. 2008;18:613–9.
303. Batra YK, Rakesh SV, Panda NB, Lokesh VC, Subramanyam R. Intrathecal clonidine decreases propofol sedation requirements during spinal anesthesia in infants. Paediatric anaesthesia. 2010;20:625–32.
304. Conklin PM. Body surface area in the infant rat. J Appl Physiol. 1975;39:335–6.
305. Shu S. Neurodiagnostic Imaging, Pediatric Hospital Medicine. In: Perkin RM, Swift JD, Newton DA, Anas NG, editors. Textbook of Inpatietn Management. 2. Philadelphia: Lippincott, Williams & Wilkins; 2008. pp. 259–268.
306. Bass NH, Lundborg P. Postnatal development of bulk flow in the cerebrospinal fluid system of the albino rat: clearance of carboxyl-(14 C)inulin after intrathecal infusion. Brain Res. 1973;52:323–32.
307. Suominen PK, Ragg PG, McKinley DF, Frawley G, But WW, Eyres RL. Intrathecal morphine provides effective and safe analgesia in children after cardiac surgery. Acta Anaesthesiol Scand. 2004;48:875–82.

Figure 1.

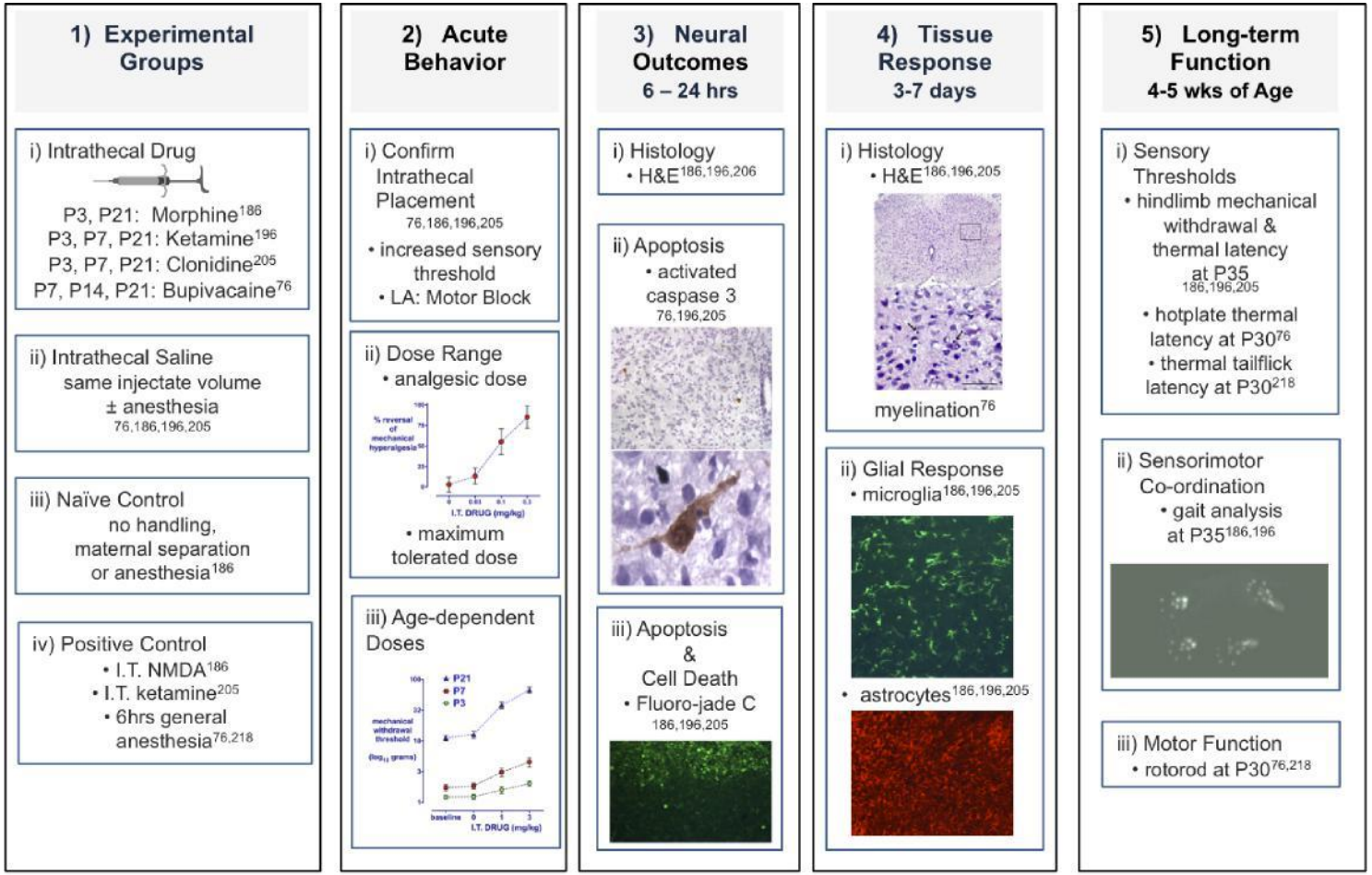
1) Experimental Groups
i) Intrathecal Drug
P3, P21: Morphine[186]
P3, P7, P21: Ketamine[196]
P3, P7, P21: Clonidine[205]
P7, P14, P21: Bupivacaine[76]
ii) Intrathecal Saline
same injectate volume
± anesthesia
76,186,196,205
iii) Naïve Control
no handling,
maternal separation
or anesthesia[186]
iv) Positive Control
• I.T. NMDA[186]
• I.T. ketamine[205]
• 6hrs general anesthesia[76,218]
2) Acute Behavior
i) Confirm Intrathecal Placement
76,186,196,205
• increased sensory threshold
• LA: Motor Block
ii) Dose Range
• analgesic dose
I.T. DRUG (mg/kg)
• maximum tolerated dose
iii) Age-dependent Doses
P21
P7
P3
mechanical withdrawal threshold
I.T. DRUG (mg/kg)
3) Neural Outcomes
6 – 24 hrs
i) Histology
• H&E[186,196,206]
ii) Apoptosis
• activated caspase 3
76,196,205
iii) Apoptosis & Cell Death
• Fluoro-jade C
186,196,205
4) Tissue Response
3-7 days
i) Histology
• H&E[186,196,205]
myelination[76]
ii) Glial Response
• microglia[186,196,205]
• astrocytes[186,196,205]
5) Long-term Function
4-5 wks of Age
i) Sensory Thresholds
• hindlimb mechanical withdrawal & thermal latency at P35
186,196,205
• hotplate thermal latency at P30[76]
• thermal tailflick latency at P30[218]
ii) Sensorimotor Co-ordination
• gait analysis at P35[186,196]
iii) Motor Function
• rotorod at P30[76,218]

Table 1 **Spinally-administered Local Anesthetics in Neonates and Infants.**

	Route	Concentration/Dose	Age	Design / sample	Outcome / Results	Side-effects/ Complications	Ref
Bupivacaine	IT	0.5mg/kg 0.5% = 0.1ml/kg (<5kg); 0.4mg/kg (>5kg)	< 1mth n=20; 1-3mths n=26; 3-6 mths n=22	Case series; inguinal hernia repair	In youngest: higher proportion (95%) withadequate spinal but less postop analgesia	Mild hypotension	288
	IT	Hyperbaric 0.5% 1mg/kg + epinephrine	Birth 24-40wks; postnatal age 5-24wks, n=20	Case series: 17 inguinal hernia, 2 stoma, 1 teratoma; BIS monitoring	100% successful block; decrease BIS values 15 mins post spinal	>20% decrease in BP; HR stable	289
	IT	Isobaric 0.5% bupi vs hyperbaric 0.5%bupi in 8%glucose;0.5mg/kg <10kg, 0.4mg/kg 11-19kg, 0.3mg/kg >20kg	2-115 mths, n=100	DB, randomized; lower abdo and lwr limb	Success (complete sensory block) higher with hyperbaric (95% vs 82%); no difference in height of sensory block (but wide variability both groups), degree motor block or postop	Treatment required: 10 suppt O_2; 1 hypotension; 1 bradycardia	290
	IT	Isobaric 0.5%; mean dose 0.68±0.16mg/kg	Gestational age at operation: 47.6 (28-1 20) wks; n=505	Case series: lwr abdo, perineal, lwr limb (79% ing hernia)	Successful spinal 96%; conversion to GA 1%(surgery >90mins); sedation required 28%	Ave. spinal tap attempts 1.4 (1-6); bloody tap 12.4%; Bradycardia 1.8% (6/9 require atropine); High block 3pts (2 req.	19
	IT + EP-B (C)	Isobaric 0.5% 1mg/kg IT in spinal grp; All: plus caudal 0.25% 2mg/kg	40 (36-44) wks PCA; n=10 (n=14 GA sevoflurane)	RCT: spinal vs GA	Successful spinal 72%; decreased postop cardiorespiratory events in spinal grp	Unsuccessful spinal 28% (4/14)	291
	IT (CSE)	IT:isobaric 1mg/kg 0.5% Caudal catheter (advanced to	31-53 wks PCA, n=28	Case series; CSE sole technique for major upr abdo surgery	Satisfactory surgical anesthesia in 24/28 (4 convert to GA); 20 supplemental midazolam;	Multiple spinal attempts 3/24; CVS parameters stable	24
	EP-B(L)	Iml/kg 0.25% bupi+epinephrine then 1 ml/kg 0.125% every 2 hrs intra-operatively	36-41 wks PCA repair on 4th±5 days (1-23dys)	Case series: bladder exstrophy repair	Intraoperative bupivacaine bolus (postop lidocaine; see below). 7/23 required intraop fentanyl.		37
Ropivacaine	IT	1.08mg/kg 0.5% = 0.216ml/kg (ED95)	<55 wks PMA n=50	Dose finding, inguinal hernia	Effective; motor block shorter duration and variable (cf other agents)		292
	EP-B EP-I	0.9-2mg/kg 0.2% (L)+ 0.2mg/kg/hr 0.2% (<6mths); 0.4mg/kg/hr	0-1mth, n=11; 1-3mths , n=10; 3-6 mths , n=10; 6-12 mths, n=14	Case series; major abdo/thoracic surgery	Decreased clearance in neonates; Unbound plasma concentration higher in neonates		293
	EP-I	0.2% 0-2ml/kg/hr	Neonate; n=22	Case series;gastro	Decreased postop ventilation in regional vs opioid		38

	EP-B(C)	1ml/kg 0.2%	0-12mths, n=10; 1-5yrs, n=10	Pharmacokinetic study; inguinal hernia	Higher plasma concentration in infants	No signs systemic toxicity	294
Levo-bupivacaine	IT	0.5 to 1.2 mg/kg 0.5%	< 55 wks PCA; n=50	Dose finding, lower abd surgery	Recommended dose 1mg/kg	No significant adverse effects	295
	EP-B(C)	2 mg/kg 0.25%	2±0.7 (0.6-2.9) months; n=22	Pharmacokinetic study; lwr abdo	Decreased clearance in infants		296
Lidocaine	EP-I	0.8-1mg/kg/hr 0.1% postoperative infusion	36-41 wks PCA repair on 4th±5 days (1-23dys)	Case series: bladder exstrophy repair *Duration: 15±8 days (4-30days)	Adjust infusion to maintain plasma concentration <5mg/L; 22/23 required reduction in infusion in first 48hrs. Tunneled catheter: 10/23 early dislodgement at 13±7 (6-28) days		37
Tetracaine	IT	Lidocaine 3mg/kg + epinephrine Tetracaine 0.4mg/kg or 0.4mg/kg + epinephrine	1mth -12 mths (7/100 < 44 wks PCA; 77/100 < 6mths); n=100	Case series: lwr abd, lwr limb (87% inguinal hernia)	Duration motor block: lido + epin 56±2.5 mins tetracaine 86±4mins tetracaine + epin 128±3mins	Four or less spinal taps attempts; no bloody taps. CVS stable.	297
	IT/ CSE	Mean dose 0.65mg/kg Post-op caudal catheter: bupivacaine 0.25mg/kg/hr (neonates) or 0.5mg/kg/hr (infants)	29wks PCA to 7 months; 1.5-7.8kg; n=19	Case series; major abdo surgery	+ CSE bupi via caudal cath	Sedation 7/19 (1 propofol, 6 midazolam); Subarachnoid block with catheter 1 (required intubation)	23
	IT	Mean dose 0.56 mg/kg 0.5% in dextrose 5%	Neonates, n=14	Case series:repair meningomyelocele	Additional doses by surgeon	Postop apnea in 2 with midazolam sedation	298
	IT	1ml/kg 0.5% tetracaine (or 0.5% bupi) + 5% glucose + adrenaline	24-42 wks gest age, n=62	Case series; 58 ing hernia, 3 pyloromyotomy, 1	Spinal success 89%;	5.4% req GA; apnea 3%; bradycardia 4%	299
	IT	0.5ml/kg 0.5% tetracaine + 5% dextrose + epinephrine	24-37 wks gest age, n=142	Case series: 95% inguinal hernia; 5% urology	Spinal success 96%;	4.5% sedation; apnea 0.8%; brady	44
	IT	Hyperbaric; mean dose 0.54±0.2mg/kg (+epinephrine in 91% (excluding PDA); [0.4% cases: hyperbaric bupi or lidocaine]	Neonates and infants (<12 mths):650g-13kg; n=1554	Case series: abdo, lwr abdo and lwr limb; urology; myelomeningocoele; (55% ing hernia)	Spinal adequate for surgery 95%;supplemental LA by surgeon 2.7%;	1.4% conversion to GA (surgery duration > block); bradycardia 1.6% (15/24 require treatment); sedation 24%; SaO2 <90% 0.6%; high block 56 patients (5 assist	13

						intubated)	
	IT	Hyperbaric 0.5% in 5% dextrose + epinephrine ; Mean dose 2.4mg/kg	Neonates mean PCA 33 (28 - 41)weeks; 1276 (650- 2965)g n=14	Case series, PDA repair	Intubated and high dose to aim for total spinal; CVS stable;	Supplement 7/14 (isoflurane, midazolam, N_2O or fentanyl);	40
Chloro-procaine	EP-B+I(C)	1ml/kg 3% ± 0.3ml/kg bolusto establish	Ex-preterm 35-49 wks PCA; n=10	Case series; feasibility in awake, inguinal	Mean cumulative requirement 2.8±1 ml/kg/hr	BP mild decrease; one apnea (pre-existing episodes)	28
	EP-B+I (C)	1-1.5ml/kg 3% bolus+ 1-1 .5 ml/kg/hr	Neonates, 1-28 days; 2.2-4.9kg; n=25	Case series, major abdo surgery (GA suppt)	CVS stable	Caudal space on 1 or 2nd attempt;	29

Legend: IT: intrathecal; EP-B: epidural bolusadministration; EP -I: epidural infusion; (L): lumbar injection; (L/T): lumbar or thoracic insertion/injection; (C): caudal injection; n.s. not statistically significant

NB: studies with LA combined with opioid or adjuvant reported in Table 2

Table 2 Spinally-administered Analgesics in Neonates and Infants (6 months)

	Route	Concentration/Dose	Age	Design / sample	Outcome / Results	Complications	Ref
Opioids							
Morphine	IT catheter	20mcg/kg bolus + 3mcg/kg/hr postop (with 0.1ml/hr 0.125% bupiv)	2-11 mths; cardiac surgery with CPB; n=30 (spinal grp)	RCT: spinal vs systemic opioid	Decreased stress response in spinal group; no difference in time to extubation	CVS stable; no spinal complications	22
	IT	7mcg/kg (with 2mg/kg tetracaine)	3mths -6yrs, n=20 spinal group	RCT:spinal vs systemic opioid	Lower pain scores and decreased postop fentanyl requirement (no diff in opioid side-effects)	CVS stable	25
	EP-B (C)	100mcg/kg (with bupi 0.25% 1ml/kg + epinephrine 1:200000)	3 – 56 mths; n=63 (31/32 per grp)	RCT: pre-incision caudal vs postop IV opioid; cardiac surgery (single ventricle	No difference in extubation rates; postop morphine reqt n.s.;	No adverse events specifically related to caudal	45
Hydro-morphone	EP-B+I	5-10mcg/kg + 0.6-1.5 mcg/kg/hr	n=220 (57 infants <12 months)	Case series cardiac surgery (range of drugs and techniques; not clear if all used in infants)	Regional safe and effective in cardiac surgery	Intravascular puncture 1; paraesthesia 7 (?age); no identifiable spinal hematoma on postop neurological	300
Dia-morphine	EP-B (C)	30mcg/kg plus bupi 0.25% 0.5ml/kg VS bupi alone	6–88 months; n=45	RCT; hypospadius repair	Reduced pain scores first 30mins postop	Minor decrease RR at 15mins; PONV n.s. difference (3/22 vs 1/23)	120
Fentanyl	EP-I	Bolus 2mcg/kg + 0.21mcg/kg/hr (+0.2mcg/kg/hr bupivacaine)	12 days -18 years n= 348 (87 <2yrs)	Case series; 80% orthopaedic	Effective analgesia	Cardiorespiratory stable; back pain at puncture site 1 (?age); fever and catheter removal 11 (tip culture all negative); mechanical problems and early cessation 25; urinary retention (without routine catheter) 17%	301
	EP-B+I (L)	1-2 mcg/kg bolus + 0.2mcg/kg/hr (plus bupivacaine 0.25% with epinephrine (up to 0.8ml/kg bolus) + 0.1% 0.2ml/kg/hr for mean 45	Neonates; n=14	Case series, major abdo surgery; mean duration 43.7±8 hrs	Satisfactory analgesia in all	Dural puncture 1/14	30
	IT	0.25, 0.5 or 1mcg/kg (plus 0.5% hyperbaric bupivacaine)	Infants, mean 6-7months; n=42	RCT; lwr abdo and urology	Addition 1mcg/kg fentanyl prolonged duration of SA block (74±6 vs 51±5 mins) and reduced postop rescue analgesia;	All low dose propofol infusion, ceased in 4 as required assisted ventilation; pruritus 3/42	302

Clonidine	IT	1mcg/kg clonidine plus hyperbaric bupi 0.5mg/kg VS 1mcg/kg fentanyl plus bupi VS 1mcg/kg clon plus fent	Infants 2-11 months (ex-preterm excluded); n=61 (15-16 per grp)	RCT DB; lwr abdo surgery under SA block (80% inguinal hernia)	Sensory block height T4-T8	Sedation score higher and intraop propofol requirement lower in BC & BCF grps; CVS stable	303
	IT	0.25, 0.5, 1 or 2mcg/kg (plus 0.5% isobaric bupi 1mg/kg)	38-46 wks PCA, n=75	RCT; inguinal hernia	Duration incr by 1 & 2mcg/kg; recommend 1mcg/kg	MAP decr by 22-40% (higher proportion MAP<40mmHg in C2); HR decr 12-27% all groups; no diff early apnea **limited follow-up until PACU discharge	26
	IT	1mcg/kg (plus 0.5% isobaric bupi 1mg/kg)	Prem vs term (29-50wks vs 39-53wks current PCA), n=67+57 = 124	Prospective observational; inguinal hernia	Success rate 84%	Unsuccessful block 10; inadequate duration 13; high block and resp impairment 1. Incr proportion apnea postop (6 before surg, 26 in 24hrs post surgery); increased	27
	EP-I (L/T)	Bolus 2mcg/kg + 0.2mcg/kg/hr OR 0.2mcg/kg/hr plus ropivacaine 0.1% (0.2mls/kg/hr)	3 - 98 months; n=35	Randomised, non blinded; major abdo surgery	"good analgesia" in both groups;; rescue analgesia required for cough and movement	Clonidine bolus increased sedation and hypotension; HR and RR stable	132
Ketamine	EP-B (C)	s-ketamine 1mg/kg ± 1 or 2mcg/kg clonidine (3groups)	1-72 mths (mean 26±24mths)	RCT; inguinal hernia	K+C longer duration; suppt analgesia in 24hrs: 63% vs 16% with combination (paracetamol? single dose)	CVS stable; "no adverse CNS effects" (?criteria); 24 hr follow-up	98
	EP-B (C)	s-ket 0.5mg/kg ± 1ml/kg levobupi 0.15% or 0.175% or 0.2%	3 mths - 6 yrs (mean 3 yrs), n=164 (52-56 per	RCT D-B; lwr abdo or urology (57% inguinal hernia)	Adequate analgesia on incision 162/164 0.175% + K: lwr analgesic reqt (22/52 vs 38/56 vs 30/56)	Postop agitation 34/164; no 'excess agitation or odd behavior'; 6hrs postop follow-up	97
	EP-B (C)	Bupi 0.125% 0.2ml/kg ± s-ketam i ne 0.5mg/kg	1 mth -9yrs (mean 2.7yrs), n=30	RCT D-B;lwr abdo or urology (60% inguinal hernia)	10/15 in ket grp vs 3/15 no additional analgesia	CVS stable, no emergence delerium or unexplained distress	137
	EP-B (C)	s-ketamine 0.5mg/kg vs 1mg/kg vs bupi0.25% 0.75ml/kg with epinephrine	3 mths -6 yrs; n=49	RCT D-B; inguinal hernia repair	ket 1mg/kg = LA > 0.5mg/kg; 33 vs 30 vs 72% suppt paracetamol	CVS stable; no difference in sedation;	144
	EP-B (C)	0.25 or 0.5 or 1mg/kg plus bupi 0.25% 0.75mls/kg	6 mths –10 yrs; n=60	RCT; unilateral inguinal hernia	0.5 and 1mg/kg prolonged analgesia and reduced rescue analgesia	1mg/kg increased behavioral side-effects 9/20 (odd, agitation,	139

						restless)	
Neo-stigmine	IT	0, 0.25, 0.5, 0.75 or 1mcg/kg neostigmine plus hyperbaric bupi 0.5%	1 –12 months; n=73 (14-15 per group)	RCT D-B; lower abdo or urology (55% inguinal hernia)	0.75 and 1mcg/kg reduced pain score and prolonged block duration;	CVS stable; emesis (10/73) did not differ across groups; apnea 6/73 (assist ventilation and cease propofol)	171
	EP-B (C)	2mcg/kg or 4mcg/kg plus levobupi 0.25% 1ml/kg vs levobupi 0.25% 1ml/kg	5 mths – 5 years; n=60	RCT: lwr abdo or inguinal (45% inguinal hernia)	2 and 4mcg/kg decrease pain score to 24hrs, prolong analgesia	PONV 4mcg/kg 3/20 (n.s.); CVS stable	162

IT: intrathecal; EP-B: epidural bolus administration; EP-I: epidural infusion; (L): lumbar injection; (L/T): lumbar or thoracic insertion/injection; (C): caudal injection; n.s. not statistically significant

Table 3: Comparison of intrathecal doses assessed in neonatal humans and rats

expressed in terms of total dose, body mass, body surface area, CSF volume and CSF turn over.					
		Analgesic Dose			
		Dosing Metric	Human Newborn	Neonatal Rat Pup (3-5 day)	
Body Measures					
	Neonatal Body weight (Kg)		3500g	12g	
	Body surface area (M*2)		Neonate = 0.2m^2 20kg=0.8; 60kg adult=1.6	0.0035m^2	
	CSF volume -mL		50	0.12	
	CSF Turn over (mcL/min)		25mls/day 17mcl/min	50mcl/day	
Analgesics			Analgesic dose	Analgesic dose	MTD or toxic dose
	Morphine	Total dose -mcg	24.5-70	0.12mcg	36
		mcg/kg	7 2 0	10	3000 (3mg/kg)
		mg/M•2	0.12–0.35	0.034	10.3
		mcg/mL CSF Vol	**0.5 –1.4**	**1 (2x)**	**300 (600x)**
		mcg per mcl/min CSF Turnover	1.44 Ð4.1	0.35	106
		Elimination mcg/ml x ml/hr	0.53–1.48	0.02	6
	Ketamine	Total dose-mcg	350	36	120
		mcg/kg	IT ~100 ¤	3000 (3mg/kg)	10,000 (10mg/kg)
		mg/M•2	1.75	10.2	3.42
		mcg/mL CSF Vol	**7**	**300 (42x)**	**1000 (142x)**
		mcg per mcl/min CSF Turnover	20.1	105	1000
		Elimination mcg/ml x ml/hr = mcg/hr	7.3	6	20
	Clonidine	Total dose-mcg	3.5	0.36	120
		mcg/kg	1	30	10,000mcg/kg 10mg/kg
		mg/M•2	0.017	0.103	3.42

		mcg/mL CSF Vol	**0.07**	**3 (42x)**	**1000 (14,285x)**
		mcg per mcl/min CSF Turnover	0.21	1.05	352
		Elimination mcg/ml x ml/hr = mcg/hr	0.07	0.06	200
	Bupivacaine	Total dose-mcg	1750 -3500	60	
		mg/kg	0.5 – 13.75 [28824] [7] [6]	*P7: 16 g	
		mg/M•2	8.75–17.5	13.3	
		mcg/mLCSF Vol	**35 - 70**	**500 (7-14x)**	
		Mcg per mcl/min CSF Turnover	102-205	176	
		Elimination mcg/ml x ml/hr = mcg/hr	37-73	10	
Reference shown in parentheses § adult analgesic doses: 0.2-0.5mg/kg epidural analgesia; 0.1mg/kg intrathecal. As 0.5mg/kg caudal epidural also analgesic in neonate,					