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Clonidine in Pediatric Anesthesia
Aspects on population pharmacokinetics, nasal administration and safety

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Abstract

Clonidine is widely used as premedication in pediatric patients and has many beneficial effects in the perioperative period. The introduction of population pharmacokinetics in the 1980s has proven useful when performing pharmacokinetic studies in children to circumvent previous limitations with traditional pharmacokinetics. The aim of the current thesis was to further study the pharmacokinetics (PK) and the pharmacodynamics (PD) of clonidine in the pediatric perioperative setting.

Population pharmacokinetics: In Study I PK-data after a clonidine bolus of 1-2 microg·kg\(^{-1}\) in 41 children were pooled with data from 4 published studies. A population PK analysis of clonidine time–concentration profiles was undertaken using nonlinear mixed effects modeling. The aim of this study was to clarify population PK in children. Clearance at birth was 3.8 l·h\(^{-1}\)·70 kg\(^{-1}\) and matured with a half-time of 25.7 weeks to reach 82% of the adult rate by 1 year of age. The relative bioavailability of epidural and rectal clonidine was unity (\(F = 1\)). In Study III the aim was to estimate the bioavailability of oral clonidine. Clonidine plasma concentrations in 8 children after oral clonidine 4 microg·kg\(^{-1}\) as premedication undergoing adenotonsillectomy were analysed. PK parameters were calculated using nonlinear effects mixed-effects models. Current data were pooled with data from 2 published intravenous studies. The oral bioavailability was found to be 55.4% (CV 6.4%; 95% CI 46.9-65.4%).

Nasal administration: In Study II the aim was to explore the absorption PK of clonidine nasal drops in children. Plasma levels from 9 children after clonidine administered as nasal drops 4 microg·kg\(^{-1}\) were analysed. Plasma PK following administration of clonidine nasal drops showed a considerable interindividual variability and absorption was delayed and limited. A nasal aerosol increases the spread of the drug in the nasal cavity, thereby optimizing the possibility for enhanced and rapid absorption as well as circumventing any possible first-pass effects that can be associated with oral drug administration. In Study IV the onset time of preoperative sedation after clonidine administered as a nasal aerosol was evaluated using a prospective, randomized, double-blind, controlled design including 60 patients receiving placebo, 3-4 microg·kg\(^{-1}\) or 7-8 microg·kg\(^{-1}\) respectively. At 45 min, adequate sedation was seen in 65% of the patients in both clonidine groups.

Safety: One of few limitations with clonidine is its association with reduced heart rate. The aim of Study V was to investigate the incidence of bradycardia in children premedicated with either oral or intravenous clonidine as compared to children not receiving pharmacologic premedication. On arrival to the operating room heart rate was recorded. 1 507 patients were included in the analysis of which 685 patients did not receive any premedication (Group 0), 305 patients received iv Clonidine (Group CIV) and 517 patients were given oral Clonidine (Group CPO). 1 in Group 0 (0.15%; 95% CI: 0-0.81%), 0 in Group CIV (0%; 95% CI: 0.00-0.98%) and 5 patients in Group CPO (0.97%; 95% CI: 0.31-2.24%) were observed to have a HR of < 85% of the 1st centile.

Conclusions: Clearance is reduced in neonates and infants. It is recommended to reduce the doses of clonidine in this age-group. Oral bioavailability of clonidine in children is reduced as compared to adults. Our results suggest that it would be necessary to administer at least twice the intravenous dose orally to get a similar effect of clonidine in children when compared with intravenous administration. The absorption of clonidine as nasal drops is low and clonidine administered as a nasal aerosol did not improve the onset time of preoperative sedation. Nasal administration of clonidine as drops or aerosol cannot be recommended if an onset time ≤ 30 min is desired. The incidence of bradycardia following premedication with clonidine in a pediatric population is very low. Hence it does not appear rational to refrain from using clonidine as premedication in children only due to the potential risk for bradycardia.
List of publications

This thesis is based upon the following papers, referred to by the Roman numerals I–V.

I. Potts AL, Larsson P, Eksborg S, Warman G, Lönnqvist PA and Anderson BJ
Clonidine disposition in children; a population analysis.
Pediatric anaesthesia 2007; 17: 924-933.

Absorption pharmacokinetics of clonidine nasal drops in children.

III. Larsson P, Nordlinder A, Bergendahl HT, Lönnqvist PA, Eksborg S, Almenrader N and Anderson BJ
Oral bioavailability of clonidine in children.

IV. Larsson P, Eksborg S and Lönnqvist PA
Onset time for pharmacologic premedication with clonidine as a nasal aerosol: a double-blind, placebo-controlled, randomized trial.

V. Larsson P, Eksborg S and Lönnqvist PA
Incidence of bradycardia at arrival to the operating room after oral or intravenous premedication with Clonidine in children.
Manuscript.
Contents

1 Introduction ........................................... 7
2 Background ........................................... 9
3 Aims .................................................... 23
4 Subjects and methods ............................... 24
5 Results .................................................. 30
6 Discussion ............................................. 36
7 Conclusion ............................................. 44
8 Acknowledgements ................................... 45
9 References ............................................. 47
List of Abbreviations

ACE  Angiotensin Converting Enzyme
ADHD  Attention Deficit Hyperactivity Disorder
ASA  American Society of Anesthesiologists
AT  Angiotensin1-receptor antagonist
AUC  Area under the curve
BSV  Between subject parameter variability
cAMP  cyclic Adenosine-Mono-Phosphate
CI  Confidence Interval
CL  Clearance
Cmax  Maximum plasma concentration
CV  Coefficient of variation
DAG  Diacyl glycerol
ECG  Electrocardiogram
EMLA™  Eutectic Mixture of Local Anesthetics
F  Bioavailability
FDA  United States Food and Drug Administration
HR  Heart Rate
IP3  Inositol trisphosphate
IV  Intravenous
MAC  Minimal Alveolar Concentration
NIBP  Non-Invasive Blood Pressure
OR  Operating rooms
PC-VPC  Prediction corrected Visual predictive check
PICU  Pediatric Intensive Care Unit
PK  Pharmacokinetic
Q  Intercompartment clearance
SD  Standard Deviation
SE  Standard Error
SpO2  Peripheral oxygen saturation
T½  Half-life
Tmax  Time to maximum concentration
Vd  Volume of distribution
VPC  Visual predictive check
Introduction

Children and infants are a susceptible population with several differences regarding physiology and psychology compared to adults. The attitude regarding clinical trials in children was previously to protect them from clinical research.\(^1\) Dosing regimens for adults were often extrapolated to pediatric populations only by weight without pediatric pharmacokinetic or pharmacodynamic data available. Efficacy, response and safety were presumed identical of those in adults but that might not be the case.\(^2\) This trend has fortunately changed to protect children through clinical research so they are exposed to clinical studied drugs with evidence based dosing regimens studied in their populations. Children have the right to benefit from scientific and pharmaceutical progress.\(^1,3\) Pharmacokinetic studies are essential for development of new drugs and optimizing dosing regimens.\(^4\) However, traditional pharmacokinetic studies are difficult to perform in the pediatric population, due to the intense sampling required. Multiple sampling requires multiple venipunctures which can be unethical.\(^5\) Especially in young infants and neonates intense sampling can be virtually impossible due to difficult venous access and limited blood volume of the patient. Population pharmacokinetics introduced in the 1980s has proven useful especially when performing drug studies in children since limited blood sampling is required. Pooling data from different studies together with the possibility to include sparse data from a larger number of individuals, as compared to traditional pharmacokinetics, will allow for better reflection of a larger variety of drug response in the study population.\(^6\) The Critical path white paper published in 2004 by the FDA (United States Food and Drug Administration) highlighted that model based drug development is essential to increase research and development of new drugs.\(^7\) Thus, various drug regulatory agencies, e.g. FDA, EMEA (European Medicines Agency) as well as many drug companies have now adopted non-linear mixed effects modeling using NONMEM as a standard model approach.\(^8\)

Premedication in children has been used since the 1940s with a variety of drugs and administration routes. The introduction of alpha-2 agonists into the field of pediatric anesthesia has put forward an alternative to the previous considered golden standard of midazolam and clonidine is widely used in Europe and Australasia in children as premedication. The European Medicines Agency and the European Union has highlighted the urgency for future trials to explore
the efficacy, safety and pharmacokinetics of clonidine in different age groups of children.9 Pioneer work regarding clonidine in pediatric anesthesia exploring pharmacokinetics and pharmacodynamic aspects was pursued by Bergendahl et al. in the 1990s highlighting many of the beneficial effects and actions of clonidine during the perioperative period. The main objective of this thesis has been to continue that work by exploring the pharmacokinetics of clonidine through a population pharmacokinetic approach, new administration routes and confirm that clonidine is a safe pharmacological premedication alternative in pediatric anesthesia.
2 Background

2.1 Historical background of clonidine

Clonidine hydrochloride is an imidazoline compound with the chemical name 2-(2,6-dichlorophenylamino)-2-imidazoline hydrochloride. Clonidine, a partial alpha-2 agonist was characterized by Wolf et al. in 1962 and was first tested as a nasal decongestant due to its alpha-1 stimulating effects and vasoconstrictive properties. However, soon its sedative, hypotensive and bradycardic effects was noted. Its mechanism of action and effects on adrenergic receptors was studied in the late 1960s and early 1970s and became thereafter primarily used as an antihypertensive drug. Only minor adverse events as dry mouth and sedation was seen in patients, during the initial period of treatment. Clonidine reduces heart rate modestly, thus, symptomatic bradycardia or orthostatic hypotension are only observed infrequently. Serious rebound hypertension following abrupt discontinuing of clonidine medication was a more severe adverse event, only seen after long-term use. This withdrawal reaction was characterized by hypertension and tachycardia associated with restlessness, insomnia, headache and nausea all due to a rebound increase of sympathetic activity. The withdrawal syndrome after cessation of clonidine for hypertension is reported in patients with a continuous treatment period of at least six days. If serious this withdrawal reaction can cause myocardial infarction and/or cerebral haemorrhage. There is no evidence for increased sympathetic activity after a single dose of clonidine. The popularity of clonidine as an antihypertensive drug decreased due to the risk for this particular withdrawal syndrome, combined with the fact that the antihypertensive effect declined with increasing plasma concentrations. Another major reason for clonidine to lose its position as an antihypertensive medication was the introduction of much more powerful drugs such as beta-blockers, calcium channel blockers and more recently ACE (Angiotensin Converting Enzym)- inhibitors and AT1 (Angiotensin1-receptor)-antagonists.

Severe rebound hypertension has also been described following anesthesia in a patient where his ordinary clonidine prescribed for hypertension had been withheld, an observation later confirmed by Kaukinen et al. However, continuing clonidine medication during and after anesthesia and surgery did not only prevent withdrawal symptoms but also stabilized the hemodynamic response during the perioperative period.
Subsequent animal studies found that clonidine administration enhance the anesthetic effect and can reduce the amount of other anesthetic drugs substantially (for both volatile agents and synthetic opioids) when using clonidine as premedication. Intravenously administered clonidine reduced halothane MAC (Minimal Alveolar Concentration) in anesthetized dogs by nearly 50%, a reduction that could be reversed by the alpha-adrenergic antagonist, tolazoline. Furthermore, Paalzow et al. were able to show that clonidine posses an analgesic effect in animals and also potentiate the analgesic effects of morphine, indicating an interaction between alpha-2 agonists and my-agonists. The morphine antagonist naloxone can not reverse the analgesic effects of clonidine indicating separate mechanisms of action.

2.2 Alpha-Adrenoceptors

The initial classification of adrenoceptors into the alpha and beta subtypes was first reported by Ahlquist 1948. Both are activated by adrenaline and noradrenaline with equal potency. The alpha-adrenoceptors were in 1977 sub-classified into alpha1- and alpha2- adrenoceptors. The alpha-adrenoceptors are transmembrane G-protein coupled receptors, with seven alpha helical segments associated with a second messenger system. The alpha1-adrenoceptors are coupled to phospholipase C, with IP3 (Inositol trisphosphate) and DAG (Diacyl glycerol) as second messengers, inducing the effects mainly by an increase of intracellular

![Diagrammatic representation of the structure of the alpha-2 adrenoceptor, G-proteins and possible effector mechanisms. The alpha-2 adrenoceptor agonist binds to the alpha-2 adrenoceptor (a2 R). This results in coupling with G-proteins due to a conformational change in the receptor protein. The alpha-2 adrenoceptor inhibits adenyl cyclase (Ac) through the inhibitory Gi protein; transmembrane signalling is mediated by the replacement of guanosine diphosphate with guanosine triphosphate. The Gi protein also activates the outward opening of a potassium (K.) channel, which results in hyperpolarisation. The Go protein is coupled in an inhibitory fashion to calcium ion (Ca2.) translocation and to the membrane-bound enzyme phospholipase C (Pc). The alpha-2 adrenoceptor is coupled through a G-protein to hydrogen (H.) and sodium (Na.) ion exchange and phospholipase A2 (PA2).]
calcium. The alpha2-adrenoceptor reduce the production of cAMP (cyclic Adenosine-mono-phosphate) through a negative coupling to adenylate cyclase inhibiting protein phosphorylation, changing ion channel conductance, which in turn results in decreased noradrenaline release. Figure 1. The inhibition of adenyl cyclase does not mediate all of the physiological effects of the alpha2-adrenoceptor. Activation of Gi-protein gated potassium channels decreases firing of cells in the central nervous system by hyperpolarisation. Alpha2-adrenoceptor stimulation can induce sodium-hydrogen-ion exchange stimulating the formation of thromboxane A2 in platelets and are also coupled to phospholipase C, with IP3 (Inositol trisphosphate) and DAG (Diacyl glycerol) as second messengers, Figure 1.

Further subdivisions into subclasses alpha2a, 2b and 2c were based on a pharmacological rather than an anatomical or functional subclassification. The alpha2-adrenoceptors are widely spread both in the central nervous system and the periphery, being identified in human brain, spleen, kidney, aorta, lung, skeletal muscle, heart and liver. Alpha2a- and alpha2c-andrenoceptors are predominantly presynaptic as opposed to alpha2b, which is mainly found postsynaptically.

Several alpha-2 agonists contain an imidazoline structure that may interact with the three imidazoline receptor subtypes identified I1-I3, of which I1 play a role in the central inhibition of sympathetic tone. Imidazoline receptors have been identified in the striatum, hippocampus and medulla oblongata in the rat brain. It has been argued that imidazoline binding sites are involved in the reduction in blood pressure caused by alpha2-imidazoline agonists. Knaus et al. showed that alpha2-abc-adrenoceptor subtype deficient mice did not decrease arterial pressure when exposed to clonidine, moxonidine or rilmenidine, indicating the role of alpha2-adrenoceptors in the antihypertensive effects of alpha-2 agonists. The alpha2a -subtype according to pharmacological and genetic alteration studies of the alpha2-adrenoceptor is likely to mediate the most common effects of the alpha-2 agonists, clonidine and dexmedetomidine.

There is a lack of substances that can selectively block or stimulate the different subtypes of the alpha2-adrenoceptors, which make them difficult to study. Subtype-specific knockout mice provide an alternative to explore the function of each adrenoceptor subtype. The antihypertensive and bradycardic effects have been found to be dependent on the alpha2a- adrenoceptor subtype. The sedative and analgesic effects by alpha-2 agonists are also likely to be transmitted through the alpha2a-adrenoceptor, since no sedative or antinoceptive effect was seen after exposure to dexmedetomidine in alpha2a(-) knockout mice. Most likely is the alpha2a-adrenoceptor involved in the anesthetic sparing effects of alpha-2 adrenoceptor agonists.
The role of alpha2b and alpha2c is less clear. The alpha2b-subtype is reported to play a role in the neurotransmission in the spinal cord and vasoconstrictive properties and salt induced hypertension. Alpha2c-subtype adrenoceptors are mainly located in the central nervous system and are indicated to have functions related to cortical arousal, which may counteract the sedative effects of alpha-2 agonists in mouse and in regulating catecholamine release from adrenal chromaffin cells. Alpha2c might be associated with peripheral hemodynamic effects, stimulating venous vasoconstriction. Physiological and pharmacological effects mediated by the various alpha2-adrenoceptor subtypes are displayed in Table 1.

**Table 1**

<table>
<thead>
<tr>
<th>Action</th>
<th>alpha2-adrenoceptor subtype</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hypotensive, bradycardiac action</td>
<td>alpha2a</td>
<td>Altman et al., MacMillan et al.</td>
</tr>
<tr>
<td>Sedative action</td>
<td>alpha2a</td>
<td>Hunter et al.</td>
</tr>
<tr>
<td>Arterial vasoconstriction</td>
<td>alpha2a and 2b</td>
<td>Links et al., Makaritsis et al.,</td>
</tr>
<tr>
<td>Venous vasoconstriction</td>
<td>alpha2c</td>
<td>Gavin et al.</td>
</tr>
<tr>
<td>Salt-induced hypertension</td>
<td>alpha2b</td>
<td>Makaritsis et al.</td>
</tr>
<tr>
<td>Antinociceptive effect</td>
<td>alpha2a</td>
<td>Hunter et al., Lakhani et al.</td>
</tr>
<tr>
<td>Presynaptic inhibition of transmitter release</td>
<td>alpha2a, 2c and 2b</td>
<td>Altman et al., Hein et al.,</td>
</tr>
<tr>
<td>Anesthetizing effect</td>
<td>alpha2a</td>
<td>Lakhani et al.</td>
</tr>
<tr>
<td>Increased spatial working memory</td>
<td>alpha2a</td>
<td>Avery et al.</td>
</tr>
<tr>
<td>Thrombus stabilization</td>
<td>alpha2a</td>
<td>Pozgajova et al.</td>
</tr>
<tr>
<td>Hypothermic effect</td>
<td>alpha2a</td>
<td>Hunter et al.</td>
</tr>
<tr>
<td>Inhibition of gastric acid secretion</td>
<td>alpha2a</td>
<td>Sallinen et al.</td>
</tr>
<tr>
<td>Inhibition of gastric motility</td>
<td>alpha2a</td>
<td>Blandizzi et al., Mullner et al.</td>
</tr>
<tr>
<td>Gastric mucosal protection</td>
<td>alpha2b</td>
<td>Fulop et al., Zadori et al.</td>
</tr>
<tr>
<td>Ion transport and fluid secretion in the small intestine</td>
<td>alpha2a</td>
<td>Gyires et al., Hildebrand et al., Liu et al.</td>
</tr>
<tr>
<td>Beneficial effects in attention deficit hyperactivity disorders</td>
<td>alpha2a and 2c</td>
<td>Levy, Cho et al.</td>
</tr>
</tbody>
</table>
2.3 Alpha-2 agonists: Pharmacodynamic effects of Clonidine and safety issues

Clonidine is an alpha-2 agonist with a reported specificity to the alpha2: alpha1-adrenoceptors of 200-220:1, exerting its main effects by stimulation of alpha2-adrenoceptors in the vasomotor center in the locus coeruleus and in the brain stem of the brain. Both these areas have a high density of noradrenergic cells and alpha2-adrenoceptors and clonidine stimulation of the alpha2-adrenoceptors inhibits noradrenaline release. Subsequently, the effects of clonidine on the vasomotor center leads to a reduction in general sympathetic tone that will lower both systemic vascular resistance and cardiac output, leading to a reduction in heart rate and systemic blood pressure.

Alpha-2 agonist stimulation has been shown to reset the baseline to which blood pressure is regulated. Thus, the baroreceptor reflex is reset at a new lower level. The baroreceptor reflex includes stretch receptors that are located in the major arteries including the aorta and carotid arteries. Increases in systemic blood pressure will stretch and thereby stimulate the baroreceptors that will produce signaling to the brainstem, which subsequently will decrease the sympathetic signaling from the brainstem to the heart and vessels. This causes reduction of heart rate, reduced myocardial contractility as well as vasodilatation, which causes normalization of the blood pressure. The baroreceptors have little or no importance on long-term blood pressure control since it easily resets to blood pressure to which it is exposed (normal reset time approximately 1-2 days).

Locus coeruleus controls wakefulness and stimulation of its alpha2-adrenoceptors results in sedation in a dose-related manner. A synergistic effect regarding sedation is seen if alpha2-adrenoceptor agonists are administered together with benzodiazepines. However, contrary to the sedation produced by benzodiazepines, alpha-2 agonists related sedation mimics ordinary sleep from which you can be awaken and adequately perform tests. In contrast to benzodiazepines alpha-2 agonists do not induce amnesia as benzodiazepines regularly do and has been reported to increase working spatial memory.

Analgesic effects of alpha-2 agonists are seen after intravenous, epidural and intrathecal administration. Analgesic effects are mediated through several modes of action but the major site of action is in the spinal cord. Central stimulation in the locus coeruleus activates descending noradrenergic antinociceptive systems originating in the brainstem suppressing nociceptive impulses in the spinal centripetal transmission. Stimulation of spinal cord located alpha2-adrenoceptors in the substantia gelatinous of the dorsal horn inhibits nociceptive signaling.

Clonidine is associated with limited and relative benign side effects and
there is no and or only minimal effects on respiration when administered in therapeu
tic doses.\textsuperscript{81} Contrary to bensodiazepins the administration of clonidine does
not potentiate the respiratory depression caused by opioids.\textsuperscript{82} Despite its benign
effects on ventilation respiratory depression and apnea has been described in cas-
es following serious intoxication.\textsuperscript{83, 84} Overall the safety profile of clonidine in
children is reassuring, since 100-1000-fold accidental overdoses has been report-
ed to be non-fatal and only rarely needing more that prolonged monitored obser-
vation in a PICU-like environment.\textsuperscript{83, 85, 86}

2.4 Clonidine and adult anesthesia

During the 1980s and 1990s several studies were published exploring the effects of
clonidine in adult anesthesia. In the preoperative setting it was found that cloni-
dine premedication produces adequate preoperative sedation, anxiolysis as well
as may have a beneficial effect on panic disorders.\textsuperscript{87-90} Intraoperatively Ghignone
et al. reported further beneficial effects of clonidine, including reduced require-
ments of induction agents, inhalational agents and synthetic opioids as well as re-
ducing the hemodynamic response of endotracheal intubation, attenuation of the
surgical stress response and improved hemodynamic stability.\textsuperscript{87, 91-93}

The use of clonidine was also found to be very beneficial in the setting of ma-
jor surgery. In patients undergoing coronary-artery by-pass grafting Kulka et al.
found that iv clonidine given as a bolus preoperative (4 microg·kg\(^{-1}\) over 30 min)
attenuated the adrenergic and hemodynamic reactions to surgical stress and in-
creased postoperative sedation.\textsuperscript{94} De Kock et al. used a similar dosing strategy
during major abdominal surgery (4 microg·kg\(^{-1}\) at induction over 20 min, fol-
lowed by an infusion of 2 microg·kg\(^{-1}\)·h\(^{-1}\) ). These authors concluded that the inci-
dence of hemodynamic events were significantly lower in the clonidine group as
compared to controls.\textsuperscript{95} Small doses of oral clonidine of 1 microg·kg\(^{-1}\) have even
been reported to reduce the incidence of intraoperatively myocardial ischemia in
patients undergoing vascular surgery.\textsuperscript{96}

Clonidine has also been shown to be highly effectively as both prophylaxis
and treatment of postoperative shivering\textsuperscript{97, 98} and can be used as a helpful adjunct
during surgery requiring controlled hypotension.\textsuperscript{99}

Clonidine has been extensively studied in the context of regional anesthesia
and has been shown to prolong and enhance both sensory and motor blockade
following neuroaxial and peripheral nerve blocks as well as reducing the need for
supplemental postoperative morphine administration.\textsuperscript{14, 100}
2.5 Premedication, Historical background

In the early days of surgery the only way to minimize the suffering for the patients was to give drugs prior to surgery. Alcohol and opium were routinely given to patients in need of amputation during the mid-1800s. In 1805 the Japanese surgeon Hanaoka described the successful use of a cocktail of alkaloids derived from the Datura Stramonium plant that he called Tsusensan. This did induce unconsciousness for several hours and was first successfully used in a woman undergoing mastectomy for breast cancer. Later in the same century nitrous oxide, ether and chloroform had been proven to have analgesic and anesthetic effects. Initially some surgeons did not use these new drugs due to the fact that they believed that pain stimulated healing. The major breakthrough using chloroform came during the civil war in America (1861-1865) when about 80 000 anesthetics with chloroform was performed. During this period no premedication was considered necessary.

In the 1860s several steps toward premedication before chloroform and ether anesthesia were taken, aiming to prevent the side effects of these early anesthetics (e.g. increased salivation, risk for vagal reflexes and hemodynamic side effects). Claude Bernard in France and Nepomuk Nussbaum in Munich discovered that administration of morphine prior to induction prolonged and intensified chloroform anesthesia. Oral chloral hydrate prior to anesthesia was introduced by Oscar Liebrich in Berlin 1869. Albert Dastre, who was working with Bernard, introduced atropine and morphine when anesthetizing dogs in order to reduce respiratory depression and vomiting caused by chloroform anesthesia. This practice rapidly became popular in continental Europe but not in Great Britain due to the perceived risk of respiratory depression.

During the era of frequent use of nitrous oxide and ether in the beginning of the 1900s Dudley Buxton published a report on using morphine, scopolamine and atropine as premedication to reduce secretions and as an adjunct anesthetic. The introduction of intravenous induction agents drastically reduced the problems with inhalational induction, ether maintenance anesthesia, and postoperative nausea and vomiting. The more frequent use of intravenous drugs for induction of anesthesia and analgesia eventually turned the trend from intramuscular premedication to oral preoperative sedatives to mainly treat anxiety during the 1960s.

In modern medical practice an ever increasing number of patients are now handled in ambulatory day-care and tight operating schedules also demand rapid turnover in operating theatres and recovery rooms. Thus, the current situation is associated with special demands regarding the timing of premedication. Another issue that may be responsible for a reduced use of premedication, is a fear for...
unwanted postoperative sedation that will delay discharge. A recent Cochrane analysis has however shown that such fears are unwarranted since there is no delay in discharged in adult patients receiving premedication compared to placebo.\textsuperscript{106}

### 2.6 Premedication in children

In the mid 1940s the need for premedication in children was emphasized, recommending morphine combined with either atropine or scopolamine, intramuscular meperidine or barbiturates administered orally or rectally.\textsuperscript{107} During 1960s commonly used drugs used for premedication was subdivided into anticholinergic, antihistamines and sedatives.\textsuperscript{108} Compounds belonging to the belladonna group was used to minimize secretions and “By paralyzing the vagal nerve endings in the heart these drugs will afford protection against cardiac slowing or syncope caused either by mechanical stimulation or by anaesthetic agents such as suxamethonium or halothane.” Antihistamines reduced vomiting and also produced sedation. If only sedation was desired pure sedative drugs were used (e.g. oral or rectal barbiturates, chloral hydrate) and if both sedation and analgesia was needed opioids (e.g. morphine and meperidine) were frequently used.

It was soon emphasized that premedication should be individualized.\textsuperscript{108} In the late 1970s the benzodiazepine midazolam was introduced in adult anesthesia\textsuperscript{109} and soon became popular also in pediatric anesthesia.\textsuperscript{110} The reason for the growing popularity of midazolam was mainly due to its rapid onset of preoperative sedation anxiolysis that allows for a smooth inhalational induction of anesthesia and since this time it has been considered the golden standard for premedication in children at many centres.\textsuperscript{111}

During recent years the use of midazolam for routine premedication has become questioned due to a number of undesirable effects associated with its use (e.g. respiratory depression if combined with opioids, the lack of any analgesic effects and induction of unwanted amnesia).\textsuperscript{112} As a result various alternatives have emerged, e.g. the use of intranasal sufentanil. Although effective in producing a rapid onset of sedation intranasal sufentanil is associated with an increased risk of respiratory depression, muscle rigidity and postoperative vomiting.\textsuperscript{113}

Ketamine is another useful alternative to midazolam and the first reports of oral ketamine for premedication in children were published in the early 1990s.\textsuperscript{114} Ketamine produces equal satisfactory sedation as midazolam\textsuperscript{115} but has an increased risk of nystagmus and vomiting.\textsuperscript{116}

Clonidine, the focus of the current dissertation, was introduced for premedication in children in the 1990s. Contrary to all other pharmacologic alternatives clonidine does not involve mental clouding but instead induce adequate preop-
erative sedation that mimics ordinary tiredness and sleep, and has no clinically relevant impact on memory or respiratory drive.\textsuperscript{117} The use of clonidine as premedication has recently been proven to be superior to benzodiazepines in a meta-analysis.\textsuperscript{118} The more selective alpha-2 agonist dexmedetomidine has emerged as an interesting alternative for premedication in children and encouraging preliminary reports using intranasal administration was published in 2008-09, showing similar or superior sedation compared to midazolam.\textsuperscript{119, 120}

If and when premedication should be used, the optimal agent and route of administration is still an ongoing debate.\textsuperscript{121} Although pharmacologic premedication of children is a useful tool to reduce preoperative stress and anxiety it does not replace adequate preoperative information (e.g. the internet-based, interactive, age-appropriate preoperative information tool Narkoswebben)\textsuperscript{122} and psychological preparation of both children and parents.

2.7 Non-pharmacological interventions used to reduce preoperative stress and anxiety

It is often stressful for both children and parents to come to the hospital for surgery. In recent years there has been a major change in the availability of information and information techniques that aim to provide better and more adequate preparation to both parents and children prior to anesthesia and surgery. The age-specific preparation of both carer and child allows them a better understanding of what will happen, what the environment looks like and insights into the risks involved. A number of information strategies are currently being used, e.g. preoperative information by an anesthetist or anesthetic nurse, web sites with virtual tours of the operating rooms and more comprehensive preoperative preparation programs. Although being an integral part of the preoperative process and an undeniable right of children and parents, there is unfortunately not much support in the literature that preoperative preparation programmes, written information pamphlets and parental and patient information are effective in preparing the child preoperatively.\textsuperscript{123}

Being admitted to the hospital, with all its new experiences, often produces a certain degree of anxiety, but the response is very individual. The need for premedication should therefore be individualized; some patients will certainly need and benefit from pharmacologic premedication, whilst many children will not.

An ongoing discussion in this context is whether parental presence at induction of anesthesia will reduce anxiety in children. A number of publications from the US show that pharmacologic premedication is superior to parental presence at induction in reducing the anxiety of the children.\textsuperscript{124, 125} This is in contrast to results in studies from other countries such as Italy, Kuwait, Thailand and Japan.
where parental presence is not only seen to reduce anxiety in both parents and child but also represents a civil right for the parent and the child.\textsuperscript{126-129} In Sweden parental presence has been routine for more than 20 years in the majority of hospitals.\textsuperscript{130} It is very rare to have any problems with the accompanying parent during the induction of anesthesia if appropriate preoperative information has been provided.\textsuperscript{131} Non-pharmacological interventions for improving anesthesia induction quality in children were recently reviewed in a Cochrane report.\textsuperscript{132} In this report a number of distraction techniques were found successful in reducing preoperative anxiety in children during induction of anesthesia, e.g. Clown doctors, handheld videogames and acupuncture.\textsuperscript{133-135}

2.8 Routes of administration for premedication in children

– current practice

Many premedications e.g. morphine and meperidine has historically been administered through the intramuscular route to provide adequate preoperative sedation for a smooth inhalational induction.\textsuperscript{108, 136, 137} However, intramuscular injections are not acceptable for an elective pediatric patient in modern practice.\textsuperscript{138} The use of intramuscular injections is only acceptable in the circulatory unstable pediatric patient in need of acute surgery using Ketamine.\textsuperscript{139} Following the introduction of topical analgesics (e.g EMLA™ and Rapydan™) for “pain-free” intravenous (IV) line placement all drugs for premedication (e.g clonidine, dexmedetomidine, midazolam, morphine) can now be administered intravenously to reduce preoperative anxiety and can be seen as a part of a balanced general anesthetic.\textsuperscript{94, 140, 141} Due to the difficulty of establishing a peripheral IV in certain children non-invasive routes are often preferred by pediatric anesthetists. Rectal administered barbiturates, chloral hydrate, ketamine, midazolam and clonidine has all been reported to produce anxiolysis and adequate preoperative sedation in children\textsuperscript{138, 142-145} but children and parents often find the rectal route distressing and its use has decreased during recent years.\textsuperscript{138} Oral administration currently represents the preferred route of administration for premedication in children scheduled for general anesthesia. Midazolam is regarded as “the golden standard” for premedication in children at many centres. Its popularity is based on its rapid onset, reduction of preoperative anxiety and inducing satisfactory sedation.\textsuperscript{111}

Midazolam has a bitter taste\textsuperscript{146} and a pronounced variable bioavailability.\textsuperscript{147} Sublingual administration has not proven to circumvent this problem\textsuperscript{138} and, thus, different additives are often used to make midazolam more palatable for children.\textsuperscript{148} Oral ketamine produce equal effective sedation as midazolam\textsuperscript{115} but has an increased risk of nystagmus and vomiting.\textsuperscript{116} Ketamine is associated with an unpleasant taste and needs to be mixed with an sweetening additive to become
acceptable for the children. Oral clonidine is tasteless and currently used as a valid drug alternative for premedication in children, inducing similar or even superior preoperative sedation compared to midazolam. Advantages with using clonidine include a number of desirable effects in the perioperative setting, e.g. reduced need for induction agent, less requirements for volatile anesthetics and peri- and postoperative opioids, as well as reduced incidence of postoperative nausea and vomiting, shivering and emergence agitation. Clonidine produces sedation that is akin to normal tiredness and sleep and is not associated with mental clouding or amnesia.

Intranasal administration of premedication has during the recent years become increasingly popular. Intranasally administrated midazolam have a more rapid onset of preoperative sedation but produces a burning/stinging in > 70% of the patients. Clonidine administered intranasally as nasal drops does not produce any discomfort but is not associated with a faster onset of action compared to oral administration. Intranasal administered dexmedetomidine at 2 microg·kg⁻¹ and sufentanil 1-1.5 microg·kg⁻¹ leads to equivalent or superior sedation than midazolam. Intranasal s-ketamine induces preoperative sedation in younger children but spill-over to the oro-hypopharynx leads to an unpleasant taste, as with oral administration. The nasal cavity in adults can only hold an administered dose of 0.2 ml per nostril, limiting the amount of drug that can be administered by the nasal route. Larger intranasal volumes will only result in a delayed and often insufficient oral administration.

2.9 Pharmacokinetics of clonidine in adults

The kinetics of clonidine has been widely explored in adults showing an oral bioavailability between 75 and 100%. It has a rapid distribution time of 10.8 ± 4.7 min but a relatively long terminal elimination half-life of 8.5 – 20 hours. Distribution volumes range between 2 and 3.42 l·kg⁻¹ and clearance rates of 3.05 -3.99 ml·kg⁻¹·min⁻¹ has been reported.

About half of the administered clonidine dose is excreted unchanged in the urine. In vitro studies report that clonidine is metabolized by the liver through a 4-hydroxylation mainly by CYP2D6, producing the main inactive metabolite p-hydroxyclonidine.

Clonidine reaches maximum plasma levels between 1-2 hours after oral administration. Onset of sedation in adult volunteers can be observed when plasma levels of clonidine reaches 0.2-2.0 ng·ml⁻¹. The antihypertensive effects are seen within the plasma concentration range of 0.4-2.0 ng·ml⁻¹, followed by a decreased antihypertensive effect with plasma levels > 2.0 ng·ml⁻¹. No decrease in blood pressure compared to baseline is seen with concentrations > 4.0 ng·ml⁻¹.
Transdermal administration has also been used reaching therapeutic levels 2 days after application and steady state within 3 days.\textsuperscript{14, 163} After epidural administration time to maximum concentration of clonidine in plasma and cerebrospinal fluid is 12 min and 31 min, respectively.\textsuperscript{79} The plasma protein binding of clonidine varies between 20-40\% in vitro.\textsuperscript{164} The kinetics of clonidine has best been described by a two-compartment model but the pharmacokinetics can be altered by entero-hepatic recirculation, which has questioned the stability of the two-compartment model.\textsuperscript{18, 160}

### 2.10 Pharmacokinetics of clonidine in children

Clinical trials regarding beneficial effects of clonidine in pediatric anesthesia were published in the late 1980s and early 1990s and initiated further studies regarding the pharmacokinetics and hemodynamic response in pediatric anesthesia. Bergendahl et al. have presented several publications regarding the pharmacokinetics of clonidine in children.\textsuperscript{165} They report a distribution volume of 0.96 l·kg\(^{-1}\) and a clearance of 4.85 ml·kg\(^{-1}\)·min\(^{-1}\) after giving different doses of clonidine after induction of anesthesia but before the start of surgery. Intravenous clonidine injection of 0.625, 1.25 and 2.5 microg·kg\(^{-1}\) resulted in a reduction of mean arterial pressure with 16, 15 and 26 \%, respectively. Seventy-five percent of the predicted reduction in blood pressure was achieved within 21 min.\textsuperscript{151, 166} After rectal administration maximum plasma concentration (C\textsubscript{max}) and the time to maximum concentration (T\textsubscript{max}) was 0.77 ng·ml\(^{-1}\) and 52 min, and after epidural administration 0.45-0.77 ng·ml\(^{-1}\) and 48-193 min, respectively. The elimination half-life after rectal administration was 12.5 h, which is comparable to adults. After rectal and epidural administration, the bioavailability of clonidine in pediatric patients is high, close to 100\%,\textsuperscript{167, 168} but no data has hitherto been available for oral administration in children. After oral clonidine administration as a lollipop Sumiya et al. in 2002 showed that patients with satisfactory sedation had higher concentrations of clonidine (0.45 ng·ml\(^{-1}\) ) compare to the group with unsatisfactory sedation (0.26 ng·ml\(^{-1}\))\textsuperscript{169}

### 2.11 Traditional Pharmacokinetics and Population Pharmacokinetics

#### 2.11.1 Traditional Pharmacokinetics

Pharmacokinetics is frequently described in compartment models with different numbers of compartments. The distribution phase is seen in the two-compartment model as contrast to the one-compartment model.\textsuperscript{6} Important pharmacokinetic parameters are: \textsuperscript{5, 6, 170}

1. **Clearance (CL), which measure the body’s capacity to eliminate a substance**
from the blood compartment.

2. Terminal half life (T½), which represents the time it takes for the body to reduce the concentration of a substance to half of its concentration in blood.

3. Volume of distribution (Vd). Is the parameter that relates the total amount of drug in the body to the concentration of a drug in plasma.

4. Area under the concentration versus time curve (AUC) describes the systemic drug exposure.

5. Maximum concentration (Cmax) and time to maximum concentration (Tmax) are extracted from concentration versus time profiles.

Blood sampling before, during and after drug administration is required to get adequate concentration versus time profiles. Intense sampling collection, accurate drug administration, accurate sampling technique and timing of sampling is important for the validity and accuracy of the pharmacokinetic parameters.5

Pharmacokinetic studies are essential for development of new drugs and optimizing dosing regimens.4 However, traditional pharmacokinetic studies are difficult to perform due to the intense sampling required. Multiple sampling requires multiple venipunctures which can be unethical since it is causing suffering for the patient and is also inconvenient for the staff.5 Especially in children and infants intense sampling can be virtually impossible due to difficult venous access and limited blood volume. The amount of blood needed for the analysis can make intense sampling a medical risk in the smallest infants. Data from traditional pharmacokinetic studies are most often generated from a healthy and homogeneous population and may, thus, not reflect the response in the patient population that later will receive the drug.

2.11.2 Population pharmacokinetics

In the 1970s therapeutic drug monitoring became increasing popular due to the possibility to adjust dosing to optimize effect and minimize toxicity. As part of this process Lewis Sheiner, University of California San Francisco, developed the use of non-linear mixed effects modeling as a way of optimizing drug dosing based on sparse data (e.g. digoxin and warfarin).171, 172 Sheiner et al. also developed the computer software NONMEM and together with his coworker Stuart Beal implemented the term Population Pharmacokinetics. However, this approach was widely criticized by bio-analytics and statisticians, who argued that it only represented a way of retrospective analysis to solve a sparse data problem of badly designed studies, which did not have adequate data regarding dosing and sample collection.8 In response Sheiner and Beal published a number of reports that emphasized the strengths of non-linear mixed effects models using the NONMEM approach,173-175 which eventually got world-wide recognition. Thus, renowned scientist from Asia, Africa and Europe, including New Zealand and
Sweden started to use this new methodology as well as contributing to its further development. Leading the development and research in Sweden is Prof. Mats O Karlsson, Uppsala University, a leading international scientist in this field, with more than 150 original articles.

The increasing costs for drug industry to produce new drugs have led to fewer new drugs being submitted to regulatory authorities. Against this background FDA, in the Critical path white paper published in 2004, has highlighted that model based drug development is essential to increase research and development of new drugs. Thus, various drug regulatory agencies, e.g. FDA (United States Food and Drug Administration), EMEA (European Medicines Agency) as well as many drug companies have now adopted non-linear mixed effects modeling using NONMEM as a standard model approach.

Population pharmacokinetics is especially useful when performing drug studies in children since limited blood sampling is required. Including sparse data from a larger number of individuals will allow for better reflection of a larger variety of drug response in the study population. Population pharmacokinetics also allows the advantage of pooling of data from different studies, which can produce a more stable pharmacokinetic analysis instead of comparing the results of small studies with different study designs. Quantifying within and in-between participant variability is included in the analysis, improving the description of the pharmacologic data. Bayesian post hoc analysis can be used to gain estimates in an individual subject. Population pharmacokinetic analysis can also be used to predict and optimize future clinical trials.

Despite the positive attributes described above the limitations of population pharmacokinetics are several:  
1. Samples representing the pharmacokinetic estimates that are analyzed must be collected.
2. Random or only trough samples may not be sufficient.
3. Errors in dosing history, in recording sampling times as well as methodological problems in drug delivery could result in biased and imprecise parameter estimates.

Furthermore, the mathematical principles are comprehensive and the time consuming analysis is in need of substantial expertise. Statistician George Box statement on experimental model design “essentially, all models are wrong, but some are useful.”

Against the background provided above further studies regarding the use of clonidine in children undergoing anesthesia and surgery are clearly warranted.
3 Aims

3.1 General aims
To further study the pharmacokinetics and the pharmacodynamics of clonidine in the pediatric perioperative setting.

3.2 Specific Aims

\textbf{Aims}
1. To clarify clonidine population pharmacokinetics in children.
2. To investigate the absorption pharmacokinetics of clonidine nasal drops in children.
3. To determine the absorption pharmacokinetics and to estimate the bioavailability of oral premedication with clonidine in children.
4. To investigate whether nasal aerosol clonidine can reduce the onset time of preoperative sedation.
5. To investigate the incidence of bradycardia in children premedicated with either oral or intravenous clonidine as compared to children not receiving pharmacologic premedication.
Subjects and methods

4.1 General

The studies were performed in accordance with the declaration of Helsinki. Study I was conducted in Auckland New Zealand, Study II in Rome, Italy, Study III-V in Stockholm and Nyköping, Sweden. All studies were approved by the local Regional Ethical Review board in Auckland, Rome and Stockholm respectively. Individual and/or parental consent were obtained in all cases when applicable.

4.2 Patient/subjects population and demographics

Study I: Pharmacokinetic (PK)-data after a clonidine bolus of 1-2 microg·kg\(^{-1}\) in 41 pediatric patients (21 male and 20 female; mean age 5.5 years range 7 days–14.8 years; mean weight 20.9 kg, range 2.8–62 kg) after pediatric cardiac surgery (Study I:I) were pooled with data from 4 previous published studies (Study I:II-I:V).

Study I:II: PK-data after a clonidine intravenous bolus of 2.5 microg·kg\(^{-1}\) in eight male ASA 1 pediatric patients (age 33 (SD 13) months; weight 15.1 (SD 3.4) kg) scheduled herniotomy surgery.\(^{166}\)

Study I:III: PK-data after a clonidine intravenous bolus of 0.625 and 1.25 microg·kg\(^{-1}\) in 16 ASA 1 pediatric patients (12 male and 4 female; age range 13–78 months; weight range 11–20 kg) presenting for minor surgery.\(^{151}\)

Study I:IV: PK-data after a rectal clonidine dose of 2.5 microg·kg\(^{-1}\) in 10 ASA 1 pediatric patients (8 male and 2 female; age 14–48 months, weight 10–20 kg) presenting for ilioinguinal hernia repair.\(^{168}\)

Study I:V: PK-data after an epidural clonidine dose of 2 microg·kg\(^{-1}\) in 8 ASA 1 pediatric patients (4 male and 4 female; mean age 1.5 years, range 1–9 years; mean weight 11.5 kg, range, 9–41 kg) undergoing ureteral reimplantation surgery.\(^{167}\)

Study II: PK-data after an intranasal clonidine dose of 4 microg·kg\(^{-1}\) in 13 ASA 1 pediatric patients (10 male and 3 female; age 22–84 months; weight 10–25 kg) scheduled for infraumbilical surgical.

Study III: PK-data after an oral clonidine dose of 4 microg·kg\(^{-1}\) in 8 ASA 1 pediatric patients (4 male and 4 female; age: 3–10.4 years; weight 10–36 kg) scheduled for adenotonsillectomy were pooled against previous published data Study I:I and I:II as described in Study I above.
Study IV: 60 pediatric patients ASA 1-2 (39 male and 21 female; age range 0.7–6.9 years weight range 10–25 kg) scheduled for outpatient surgery or procedure. 
Study V: 1 507 pediatric patients (899 male and 608 female; age range 0.02-18 years) scheduled for anesthesia.

4.3 Anesthetic protocol

Study II: Patients received 5 mg·kg⁻¹ of oral ibuprofen 30 min prior to an inhalational induction with sevoflurane and nitrous oxide. Two intravenous catheters were placed, one was used for blood sampling only. Anesthesia was maintained with sevoflurane in a mixture of oxygen and air. All patients were breathing spontaneously through a laryngeal mask airway. Caudal or peripheral nerve block was used according to surgical procedure. A dose of 4 microg·kg⁻¹ of clonidine was then administered intranasally divided up in equal volumes for both nostrils with the patient in supine position and head in neutral position for the entire length of surgery.

Study III: All patients received rectal paracetamol 40 mg·kg⁻¹ and rectal ibuprofen (62.5 or 125 mg depending on body weight). Following the application of EMLA™ cream to one upper extremity, an intravenous catheter was inserted for the purpose of blood sampling as well as for subsequent intravenous anesthesia induction. Following this, the patients were given 0.1 ml·kg⁻¹ of an extemporaneous solution of clonidine 40 microg·ml⁻¹ and atropine 20 microg·kg⁻¹ mixed with 30 ml of apple fruit drink, administered orally. This oral premedication dose was given at least 1 h prior to the planned anesthesia induction. Induction of anesthesia was performed with fentanyl 1 microg·kg⁻¹ and propofol 2-4 mg·kg⁻¹. Endotracheal intubation was facilitated by the administration of suxamethonium 1 mg·kg⁻¹ and anesthesia was maintained by sevoflurane in 30% oxygen. A throat pack was inserted immediately after endotracheal intubation to prevent the passage of blood into the stomach. No gastric emptying or gastric suctioning was performed that might influence the gastrointestinal absorption of clonidine.

Study IV: Patients were randomized to receive saline placebo (group P), clonidine 3–4 microg·kg⁻¹ (Group C4), or clonidine 7–8 microg·kg⁻¹ (Group C7). The Hospital pharmacy prepared the nasal spray using a 600 microg·ml⁻¹ clonidine hydrochloride base solution that was adequately diluted with saline according to patient weight and randomized dose. The nasal pump administration device delivers an aerosol with a volume of 100 microl per spray. According to the weight of the patients, two or three sprays of the study solution were finally administered to the patients, divided between the two nostrils. After preoperative evaluation anesthesia was induced with fentanyl 0.5–1.0 microg·kg⁻¹ and propofol 2–4 mg·kg⁻¹. A laryngeal mask airway was inserted or endotracheal intubation was
performed using suxamethonium or rocuronium to facilitate intubation. Anesthesia was thereafter maintained with sevoflurane in oxygen and air. Regional anesthetic blocks were used when applicable. Paracetamol, intravenous ketorolac, and/or morphine were used for postoperative analgesia.

4.4 Blood sampling and plasma concentration measurements

4.4.1 Blood sampling

**Study I:** Blood samples for clonidine assay were taken at 5 min, 3 and 8 h following administration and collected into a heparinized test tube. The plasma was separated by centrifugation and stored at -80 °C until assay.

**Study II:** Blood samples for clonidine assay were taken at 5, 10, 15, 30, 60, 120, 180 and 360 min following administration and collected into a heparinized test tube. The plasma was separated by centrifugation and stored at -30 °C until assay.

**Study III:** Blood samples for clonidine assay were taken at 15 and 30 min following administration and collected into a heparinized test tube. The plasma was separated by centrifugation and stored at -30 °C until assay.

**Study IV:** Blood samples for clonidine assay were taken at 5, 10, 15, 20, 25, 30, 45 min and 1, 2, 3, 6, 12, 18 and 24 h following administration and collected into a heparinized test tube. The plasma was separated by centrifugation and stored at -20 °C until assay.

**Study V:** Blood samples for clonidine assay were taken at 2, 5, 15, 30, 45 min and 1, 3, 6 and 10 h following administration and collected into a heparinized test tube. The plasma was separated by centrifugation and stored at -20 °C until assay.

**Study VI:** Blood samples for clonidine assays were taken at 10, 25, 40, 60, and 90 min and 3, 6, and 12 h following administration and collected into a heparinized test tube. The plasma was separated by centrifugation and stored at -70 °C until assay.

**Study VII:** Blood samples for clonidine assays were taken immediately before administration, at 5, 15, 30, 45 min, and 1, 2, 4, 6, 12 and 18 h following administration and collected into a heparinized test tube. The plasma was separated by centrifugation and stored at -70 °C until assay.

4.4.2 Clonidine assays

**Study I:** The analysis was performed on a mass spectrometer with Turbolon-Spray, ion source operating in positive ion mode linked to a liquid chromatography system. The lower limit of quantification was 0.1 ng·ml⁻¹. The interassay coefficient of variation (CV) was 3.71–11.5% and the intra-assay CV was 3.35–14.7%.

**Study II-V:** Clonidine analysis was performed by radioimmunoassay. The detection limit of the assay was 0.1 ng·ml⁻¹ At reference concentrations 0.20, 0.50 and 1
ng·ml⁻¹, the deviation of the found values were 0, -2 and -9% of the reference values, respectively. Inter- and intra-assay coefficients of variation (CV) were 5–8% and 4–5%, respectively.

**Study II and III:** The clonidine analysis was performed on an ultrapressure liquid chromatography coupled to a mass spectrometer. The detection limit of the assay was 0.1 ng·ml⁻¹. The inter assay coefficient of variation (CV) was 6.0% and the intra assay CV was 5.2% for the low concentration quality control. The inter-assay CV was 5.2% and the intra assay CV was 4.1% for the high concentration quality control.

### 4.5 Pharmacokinetic analysis

**Study I:** A two-compartment (central and peripheral) linear model fitted data better than a one-compartment pharmacokinetic model. An additional depot compartment was used to characterize absorption from the epidural and rectal sites. Population parameter estimates were obtained using nonlinear mixed effects modeling (NONMEM). The population parameter variability in model parameters was modeled by a proportional variance model. An additive and a proportional term were used to characterize the residual unknown variability. A separate set of residual errors was used to distinguish assay data from Auckland (cardiac surgery) from those from Stockholm (general surgery). The population mean parameters, between subject variance and residual variance were estimated using the first-order conditional interaction estimate method using ADVAN4 TRANS4 of NONMEM V. The population parameter variability is modeled in terms of random effect (η) variables. The covariance between two elements of η (e.g. CL and V) is a measure of statistical association between these two variables. The covariance of clearance and distribution volume variability was incorporated into the model. The parameter values were standardized for a body weight of 70 kg using an allometric scaling model. We anticipated that children after cardiac surgery may have altered clearances and volumes of distribution compared with those children undergoing general surgery. Scaling factors were applied to these CL and V in those children undergoing cardiac surgery. The scaling factor applied to V1 was FV1, while that applied to V2 was FV2. The quality of fit of the pharmacokinetic model to the data was sought by NONMEM’s objective function and by visual examination of plots of observed vs predicted concentrations. Models were nested and an improvement in the objective function was referred to the Chi-squared distribution to assess significance e.g. an objective function change of 3.84 is significant at α = 0.05.

**Study II:** The PC NONLIN program (version 2.0, Statistical Consultants Inc, Lexington, Kentucky, USA) was used for the pharmacokinetic modeling of the plasma concentration data. The maximum plasma concentration (Cmax) and
time to achieve maximum plasma concentration (Tmax) were evaluated from the fitted curves. The area under the curve (AUC) was calculated by numeric integration of the plasma concentration–time (12 h) curve for each patient.

Study III: A two-compartment (central and peripheral) linear model was used to fit data. An additional depot compartment was used to characterize absorption from gut. Population parameter estimates were obtained using nonlinear mixed-effects models (NONMEM VI; Globomax LLC, Hanover, MD, USA). The population parameter variability in model parameters was modeled by a proportional variance model. A proportional term (Err) was used to characterize the residual unknown variability. These population mean parameters, between-subject variance and residual variance, were estimated using the first order conditional interaction estimate method using ADVAN4 TRANS4 of NONMEM VI.

The population parameter variability is modeled in terms of random effect (η) variables. The covariance between two elements of η (e.g. CL and V) is a measure of statistical association between these two variables. The covariance of clearance and distribution volume variability was incorporated into the model. Each study was initially assigned an individual residual error. The parameter values were standardized for a body weight of 70 kg using an allometric scaling model. Bootstrap methods, incorporated within the Wings for NONMEM program, provided a means to evaluate parameter uncertainty. A total of 1000 replications were used to estimate parameter confidence intervals. A visual predictive check (VPC) was used to evaluate how well the model predicted the distribution of observed clonidine concentrations. Simulation was performed using 1000 subjects with characteristics taken from studied patients. For data such as these where covariates such as dose, weight, height and sex are different for each patient, we used a prediction corrected VPC (PC-VPC). Observations and simulations are multiplied by the population baseline value divided by the individual estimated baseline.

4.6 Pre- and postoperative assessment

Study IV: Immediately prior to the administration of the nasal aerosol, the following baseline data were registered: sedation score, heart rate (HR), peripheral oxygen saturation (SpO2), and noninvasive systolic blood pressure (NIBP). Acceptance of the nasal aerosol was assessed either as well accepted (no or only minor reaction to the administration) or poorly accepted (major adverse reaction or crying). Following the administration of the study drug, patients were assessed every 5 min for 45 min. Assessment of sedation was performed using The Children’s Hospital of Wisconsin Sedation scale.178

The scale is graded 0–6 (0 unresponsive, 1 arouses but not to consciousness with painful stimulus, 2 arouses slowly to consciousness with sustained painful stim-
ulus, 3 arouses to consciousness with moderate tactile or loud verbal stimulus, 4 drowsy, eyes open or closed, but easily arouses to consciousness with verbal stimulus, 5 spontaneously awake without stimulus, and 6 anxious, agitated, or in pain) with satisfactory sedation scores defined as score 3 or 4. Heart rate and SpO2 were assessed by pulse oximetry at 20 and 40 min after administration. Postoperative sedation was assessed by the recovery room nurses using a modified Ghai scale$^{149}$ (1 = awake, calm quiet, 2 = awake but tired, 3 = drowsy but responds to verbal commands or gentle stimulation, 4 = asleep, and 5 = anxious, depressed/agitated/crying) with satisfactory scores defined as <4.

4.7 Registration of Heart rate and definition of reduced Heart Rate

*Study V:* On arrival to the operating room (OR) the initial heart rate (HR) was recorded following the patient being connected to either pulseoximetry or electrocardiografic monitoring (ECG). Data from Fleming et al.$^{179}$ were used to determine normal heart rates according to age. In accordance with the chosen reference for normal pediatric heart rates, values below the 1st centile in the total study population were defined as bradycardia (BradyT). Since most anesthetist will not treat HR values just below the normal lower HR limit we arbitrarily defined a HR below 85 % of the lower limit of the normal range (1st centile)$^{179}$ as bradycardia that might need clinical intervention (BradyCI). (e.g. IV atropine or glycopyrrolate).

4.8 Statistical Analyses

*Study I and III:* The statistical analyses are incorporated as a part of the population pharmacokinetic model analyses as described above.

*Study II:* All numerical data are reported as median. Calculation of nonparametric 95% confidence intervals (CI) were based on Wilcoxon signed ranks test.

*Study IV:* Classified data from two independent populations were compared by the Fisher’s exact test. Classified data from several independent populations were compared by the chi-squared test for independence. Non-classified data from the three groups were compared by the Kruskal–Wallis test. P-value < 0.05 was considered as statistically significant. All reported P-values were from two-sided tests.

*Study V:* The Kruskal–Wallis analysis (non-parametric ANOVA) with the Dunn’s post hoc test was performed for multiple comparisons of independent groups of samples. The 95% confidence intervals for proportions were calculated as given in.$^{180}$ Classified data from two independent populations were compared by the Fischer exact test. P-values less than 0.05 were considered as statistically significant. All reported p-values were from two-sided tests.
5 Results

5.1 Included/Excluded patients
A total of 1,676 patients were included. No exclusion of patients in Study I and V. Four patients were excluded in Study II, five patients in Study III and seven patients in Study IV.

5.2 Clonidine disposition in children (Study I)
The analysis included 380 observations from 72 children. Population parameter estimates (between subject variability) were clearance (CL), central volume of distribution (V1), intercompartment clearance (Q) and peripheral volume of distribution standardized to a 70 kg person, displayed in Table 2. Clearance at birth was 3.8 l·h⁻¹·70 kg⁻¹ and matured with a half-time of 25.7 weeks to reach 82% adult rate by 1 year of age, Figure 2. The relative bioavailability of epidural and rectal clonidine was unity (F = 1).

<table>
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<th>Parameter</th>
<th>Estimate</th>
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<th>%SE</th>
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<td>V1</td>
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</table>

Table 2
Standardized clonidine population pharmacokinetic parameter estimates (BSV is the between subject parameter variability, SE is the standard error of the estimate). (b) Alternative parameterization. (c) Covariate analysis (BSV is the between subject parameter variability, SE is the standard error of the estimate)

CL = (CLstd [70]⁻⁰·⁷³) \(\left((1-(1-\beta_d))e^{-\text{PNA}\frac{0.23}{2}}\right)\) l·h⁻¹, where CLstd is the population estimates for CL, standardized to a 70-kg person using allometric models; PNA is the postnatal age in weeks; \(\beta_d\) is a parameter estimating the fractional difference from CLstd at birth; \(T_d\) describes the maturation half-life of the age related changes of CL. FV1 and FV2 are scaling factors applied to V1 and V2 in children who had cardiac surgery.
Figure 2
Individual predicted clonidine clearances (CL), standarized to a 70 kg person, from the NONMEM posthoc step, are plotted against postnatal age. The solid line represents the nonlinear relation between clearance and age.

5.3 Pharmacokinetics of clonidine nasal drops (Study II)

The plasma absorption PK of clonidine nasal drops 4 microg·kg^{-1} was slow and modest and showed a considerable interindividual variability, Figure 3. The median Cmax was 0.53 ng·ml^{-1} (95% CI 0.42–0.64 ng·ml^{-1}) and the median Tmax was 2.22 h (95%CI 1.45–2.99 h). The median AUC for data from 0 to 12 h was found to be 7.24 ng·ml^{-1} (95% CI 5.63–8.85 ng·ml^{-1}) The median elimination half-life time (K10-HL) was 7.69 h (95% CI 5.22–10.16 h). Thus the PK of clonidine administered as nasal drops showed a substantial variability.

Figure 3
Plasma pharmacokinetics (PK) of nasal clonidine in children. Plasma concentration – time curves of nine patients are included.
5.4 Oral bioavailability of clonidine (Study III)

The median oral bioavailability was 55.4% (CV 6.4%; 95% CI 46.9-65.4%). The plasma time–concentration profiles are displayed in Figure 4 and population parameter estimates are shown in Table 3.

![Plasma concentration over time](image)

**Figure 4**
Individual plasma concentration time data from all patients participating in the study. Plasma concentration of clonidine that has been estimated as satisfactory for preoperative sedation in children 0.3 ng·mL⁻¹ is indicated by the dotted line.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Estimate</th>
<th>% BSV</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>$CL_{\text{std}}$ (L·h per 70 kg)</td>
<td>17.9</td>
<td>30.3</td>
<td>16, 20.3</td>
</tr>
<tr>
<td>$V_{1\text{std}}$ (L·70 kg⁻¹)</td>
<td>81.2</td>
<td>71.5</td>
<td>60.7, 105</td>
</tr>
<tr>
<td>$Q_{\text{std}}$ (L·h per 70 kg)</td>
<td>121</td>
<td>44.3</td>
<td>80.1, 165</td>
</tr>
<tr>
<td>$V_{2\text{std}}$ (L·70 kg⁻¹)</td>
<td>113</td>
<td>33.9</td>
<td>91, 131</td>
</tr>
<tr>
<td>Tabs (h)</td>
<td>0.454</td>
<td>85.1</td>
<td>0.221, 0.884</td>
</tr>
<tr>
<td>Tlag</td>
<td>0.148</td>
<td>91.2</td>
<td>0.002, 0.316</td>
</tr>
<tr>
<td>$TM_{50}$ (weeks)</td>
<td>61.6</td>
<td>–</td>
<td>53, 78.8</td>
</tr>
<tr>
<td>N</td>
<td>2.42</td>
<td>–</td>
<td>1.5, 3.72</td>
</tr>
<tr>
<td>F with oral</td>
<td>0.554</td>
<td>6.4</td>
<td>0.469, 0.654</td>
</tr>
<tr>
<td>Err</td>
<td>0.132</td>
<td>–</td>
<td>0.105, 0.155</td>
</tr>
</tbody>
</table>

**Table 3**
Standardized clonidine population pharmacokinetic parameter estimates (BSV is the between-subject parameter variability, CI is the confidence interval.)
5.5  Premedication with clonidine as a nasal aerosol
(Study IV)

In the group receiving 7-8 microg·kg\(^{-1}\) of clonidine, 55% of the children were found adequately sedated at 30 min as compared to 32% in the group receiving 3-4 microg·kg\(^{-1}\) (\(P = 0.1202\)), and 9.5% in the placebo group (\(P = 0.0025\)), Figure 5. At 45 min postadministration, adequate sedation was seen in 65% of the patients in both groups receiving clonidine, which were both found to be significantly higher compared with the placebo control group (14%; \(P\)-values = 0.0027 and 0.0013, respectively), Figure 5. During the 2-h postoperative observation period, the sedation profile did not differ between the three study groups, Figure 6.

![Figure 5 Percentage of patients with adequate sedation during the 45-min observation period postadministration. ○ = Placebo group ■ = Group C4 , ▽ = Group C7.](image)

![Figure 6 Sedation profile during the 2-h postoperative period. ○ = Placebo group ■ = Group C4 , ▽ = Group C7.](image)
5.6 Incidence of bradycardia (Study V)

Data from 1,507 patients were collected and included in the analysis. 685 patients did not receive any pharmacologic premedication (Group 0), 305 patients received intravenous Clonidine (Group CIV) and 517 patients were given oral Clonidine (Group CPO). The median (range) HR on arrival to the operating rooms (OR) for Group 0, CIV and CPO were 98 (45-190) bpm, 90 (46-172) bpm and 93 (40-189) bpm, respectively. HR was significantly lower in Group CPO and CIV in comparison to Group 0 (p values < 0.01).

Ten patients in Group 0, 6 in Group CIV and 26 in Group CPO were classified as BradyT (a HR below the 1st centile) upon arrival to the OR, Figure 7. The incidence of BradyT was significantly higher in Group CPO in comparison to Group 0 (p values < 0.01), Figure 8. One patient in Group 0, 0 in Group CIV and 5 patients in Group CPO fulfilled the criteria for BradyCI (a HR of < 85% of the 1st centile) upon arrival to the OR, Figure 7.

No statistical differences in the incidence of BradyCI were seen between the groups (p-value < 0.05), Figure 8. In BradyCI patients atropine was administered before induction to the 1 patient in Group 0 and in 2 out of the 5 cases in Group CPO, resulting in an appropriate increase in HR. In the remaining three BradyCI cases the attending anesthetist opted to accept the observed HR.

![Heart rate on arrival to OR](image)

**Figure 7**
*Heart rate on arrival to OR*
- ▼ = Group 0, Figure 7A
- ○ = Group CIV, Figure 7B
- ● = Group CPO, Figure 7C
- — = lower limit of the normal range (1st centile)
- —— = 85% of the lower limit of the normal range (1st centile)
Figure 8
Incidence of patients with a reduction of HR on arrival to OR below the 1st centile (A) and 85% of the 1st centile (B) in Group CPO, CIV and 0 respectively with 95% Confidence interval.
6 Discussion

6.1 Population pharmacokinetics and oral bioavailability (Study I and III)

Traditional pharmakocinetic (PK) studies are important in the descriptions of PK-data in individuals for regulatory and research purposes were the data describes PK-data with a strong individual accuracy but depend on high numbers of samples in each individual and may not reflect the sub-populations of patients that might frequently be exposed to the drug. In the adult population were blood sampling is not an issue there is a place for traditional PK analysis but in pediatric studies it is questionable to do intense sampling. Individual PK-analysis with intense sampling in patients in need of cytotoxic agents with a narrow therapeutic index can benefit from a PK-analysis to clarify the PK-response in that specific individual. The possibility of using sparse sampling technique and unbalanced data sets is optimal in pediatric studies for ethical reasons due to discomfort pain and anxiety associated with venipuncture and the amount of blood samples that can be obtained.6

The population pharmacokinetic model is built using three parts. A structural (fixed effects), statistical (random effects) and covariate model are chosen. These three parts describes the trends in the data, intersubject-, intrasubject- and residual variability and relationships between covariates (e.g. weight and age) and parameters of the structural model, respectively. Finding a model that describes the data adequate requires model checking and refining in several steps. Scatter plots or goodness-of-fits-plots are used to assess model fit in relation to observed concentrations. The basic internal model evaluation is part of the final stage of the model building procedure to assess whether the model is able to describe the data set that was used to develop the model accurate and without bias.

Finally an external evaluation is performed to assess if the model can describe an external dataset adequately, a dataset that was not used to develop the model. There are various techniques used for both internal and external validation (e.g. Basic Goodness of-fits-plots, Bootstrap analysis, Visual predictive checks). If the model performs well, simulating concentrations and/or effects and their variability can be predicted when different doses are tested the dosing regim that results from the model can be tested in a clinical trial.181
It is important to collect samples representing PK-parameters that are analyzed since random or only trough samples may not be sufficient. In Study I plasma samples from the cardiac patients were sampled at 5min, 3h and 8h after administration. With a distribution half life of 12 min the study population could have been divided in two group with different sampling schemes (Group 1 at 5, 15 and 120 min and Group 2 at 10, 30, 480 min following administration) to better describe the initial part of the plasma concentration profile. Assays performance, with a high but acceptable coefficient of variation can differ between studies when pooling data from different studies. In Study I a separate set of residual errors was used to distinguish assay data from the cardiac patients from the general surgery patients.

The data from Study I has made it possible to perform simulations exploring the plasma concentration profiles in different age groups exposed to different doses of clonidine. These simulations have beneficially contributed to the dosing regimen in the upcoming CloSed trial (personal communication Joe Standing), a major EU 7th Framework sponsored study (EU contribution ~6,0 million euro), which represents a multicenter double-blind randomized controlled trial comparing clonidine and midazolam as infusions for sedation in the PICU environment, http://www.closed-fp7.eu/.

Study III was planned as traditional PK-study with intense sampling in each individual. Samples from 13 patients were analyzed 8 patient fitted the model accurate in the first traditional analysis with a two-step approach and was planned to described oral bioavailability in clonidine comparing oral plasma concentration profiles with previously published iv plasma concentration profiles using a previous methodology that had been used in two published studies describing rectal and epidural bioavailability of clonidine but without population pharmacokinetics. The traditional PK-analysis with this approached failed to display a robust model with reliable data on oral bioavailability. Using the PK-data that we knew fitted the traditional PK-model an attempt was made to apply a population-PK analysis to the 8 patients included in the traditional PK-analysis. A robust model was achieved and described for the plasma concentration profiles of the patients included in Study III. Estimates of Tabs and F are sometimes erroneous with all approaches except for paired analysis by comparison of the area under the plasma concentration vs time curve (AUC) after drug administration by each of the enteral and intravenous routes in the same individual, the current methodology is acceptable. Only 8 patients were included in the final analysis. Populations PK-analysis accepts sparse data and adjusts for covariates as age and weight. The excluded patients with sparse data that did not fit the primary traditional PK-model could have been tested and included in the final analysis. Results
from population pharmacokinetic studies from sparse samples in a small number of subject can generate unreliable parameter estimates. Our results suggest that it would be necessary to administer at least twice the intravenous dose orally to get a similar effect of clonidine in children when compared with intravenous administration. This is in agreement with our underlying observation that 4–5 microg·kg⁻¹ is needed for oral premedication, whereas only 2.5 microg·kg⁻¹ is necessary after rectal administration. The current study design is unable to explain the observed reduced relative bioavailability of the oral formulation in children compared to adults. Diluted apple fruit juice was used as a flavored carrier. Since fruit juices and constituents can reduce bioavailability drug absorption, the supposition is that the apple juice carrier could impact on clonidine absorption is speculative.

6.2 Nasal administration and absorption pharmacokinetics (Study II and IV)

There are several reports regarding the efficacy of Clonidine as premedication when administered through the oral or rectal route producing effective preoperative sedation even superior to the effect achieved by benzodiazepines in children. However, the onset of action by the oral route can take as long as 105 min. Intranasal administration is a practical, non-invasive, rapid, and simple method. The highly vascularized nasal mucosa with its large surface area enhances both drug absorption and the onset of therapeutic action. Lipophilic drugs are generally well absorbed when administered intranasally with high bioavailability. A good example of this is intranasal administration of fentanyl where the time to maximum concentration is fast, 7 minutes or less, and similar to intravenous administration. The reported bioavailability of intranasally administered fentanyl is 80%. A study on rodents reported that nasal administered clonidine could be an alternative to intravenous administration as Cmax were reached after 10 min. Almenrader et al. has previously demonstrated that clonidine administered orally and nasally as premedication is well tolerated by children and with no significant difference of the onset time for preoperative sedation between the groups. Intranasal fentanyl, administered after induction of anesthesia in children, does result in adequate and timely plasma levels. Our aim in Study II was to investigate the absorption pharmacokinetics of clonidine administered as nasal drops. The plasma concentrations in this study were found to be within range of plasma concentrations associated with clinical effects regarding sedation in children (0.3 ng·ml⁻¹). Plasma concentrations were lower than those found after rectal or epidural clonidine administration. Time to achieve maximum plasma concentration was approximately 2 h, i.e. 20 times longer than in the
rodent study\textsuperscript{187} and more than twice as long as the time to achieve the maximum sedative effect in our previous clinical study.\textsuperscript{155} The administration of nasal drops in an anesthetized child differs from that of the awake child who is usually in a sitting position and breathing partly through the nose. Together with the supine position this may have increased the risk that a substantial part of the administered dose would have been a delayed oral administration. Due to the unexpected absorption pattern of clonidine as nasal drops after induction of anesthesia the number of blood sampling time points was limited at the time when $C_{\text{max}}$ and $T_{\text{max}}$ values actually did occur. This does render our estimates of $C_{\text{max}}$ and $T_{\text{max}}$ somewhat imprecise but does not alter the conclusion that nasal administration as nasal drops cannot be recommended due to the great interindividual variability of absorption associated with this administration technique.

The nasal route is very accessible and frequently used for drug administration for painful procedures in children\textsuperscript{156} and many children have previously also been exposed to nasal decongestants. This together with the fact that clonidine administered orally or as nasal drops in children induces adequate preoperative sedation and also possesses many beneficial effects in the perioperative period led to Study IV.

The onset of action of clonidine is slow after oral or nasal drop administration.\textsuperscript{155} However, delivery of drugs as a nasal aerosol increases the spread of the drug in the nasal cavity,\textsuperscript{189} thereby optimizing the possibility for enhanced and rapid absorption as well as circumventing any possible first-pass effects that can be associated with oral drug administration.\textsuperscript{190} Thus, nasal aerosol administration is often associated with a more rapid onset time compared with oral or nasal drop administration.\textsuperscript{189, 191} The primary goal of Study IV was to potentially shorten the often prolonged onset time of sedation associated with the use of clonidine administered orally or as nasal drops\textsuperscript{155, 184, 192} to a clinically more acceptable period of $\leq 30$ min by administering clonidine as a nasal aerosol. Unfortunately we were unable to reduce the onset time to $\leq 30$ min even when using a quite high total dose of clonidine (7–8 mikrog$\cdot$kg$^{-1}$). Even at the end of our 45-min observation period, only 65% of the children given clonidine as a nasal aerosol achieved the desired level of preoperative sedation. This may be explained by at least four different reasons. First the amount of drug the pediatric nasal mucosa might be smaller than the 100-200 mikrol per nostril reported in adults. If so, there could be a potential risk that a part of the dose would run off to the oropharynx as a delayed oral administration, despite concentrating the solution leading to a spray volume of only 100-200 mikrol per nostril. A second possible mechanism for a delayed effect of nasal aerosol clonidine could be an alpha-1 adrenoceptor-mediated vasoconstriction in the nasal mucosa that may reduce systemic uptake.
Third, in-vitro findings, clonidine requires a specific transportation system to pass through the endothelium of cerebral microvessels. Thus, clonidine has a slower transendothelial transport across the blood–brain barrier than could be expected from its high degree of lipophilicity. A possible fourth mechanism could be due to the fact that we included patients for more than a year even during the winter seasons and not excluding patients with a minor runny nose that was accepted by the attending anesthetist. An increase of nasal secretion can lead to a reduction of drug uptake and subsequent decreased drug effect.

Despite high doses of clonidine up to 7–8 mikrog-kg\(^{-1}\) there were only two patients that had brief episodes of relative bradycardia and none of these patients needed any additional treatment other than light stimulation to resolve the problem. However, our observations are well in line with the published literature where major hemodynamic side effects of clonidine appear rare if the administered dose is < 10 mikrog-kg\(^{-1}\). The number of patients in Study IV is too limited to allow any relevant overall safety statement.

**6.3 Aspects on safety and incidence of bradycardia (Study V)**

Clonidine is associated with a high degree of safety in children, with 100-1000-fold accidental overdoses reported to be non-fatal, and has a limited and relative benign side-effects spectrum. However, a reduction in heart rate is one side effect that is frequently put forward as an argument for hesitating to adopt clonidine as a routine agent for premedication in children, the fear being that the reduction in heart rate may progress to frank bradycardia that may lead to serious hemodynamic compromise. Based on our now long-standing clinical experience of using clonidine as the preferred standard choice of pharmacologic premedication in all age groups, combined with the reported safety of substantial accidental overdoses we believe that such fears are unwarranted and does not represent a valid argument for refraining from use of clonidine as premedication in children.

Thus, the aim of Study V was to investigate the incidence of bradycardia in children premedicated with either oral or intravenous clonidine as compared to children not receiving pharmacologic premedication. Patients premedicated with clondine displayed a reduction in HR but only few children were found to have heart rates below the arbitrary definition for Brady CI (< 85 % of the 1st centile). Oral clonidine is recommended to be administered 45 min - 105 min before expected induction of anesthesia to optimise preoperative sedation when using oral clonidine, to stimulate Growth Hormone, a HR reduction reached a nadir of on average 8.4% compared to baseline 2-3 h after administration.

This may explain the low heart rates in two BradyCI patients who received
oral clonidine 3.4 h and 4.4 h before arrival to OR, respectively.

HR below the lower normal limit for age after clonidine premedication usually responds well to only minor-moderate vocal or cutaneous stimulation. The decision to administer atropine or glycopyrrolate to patients classified as BradyCI was left at the discretion of the attending anesthetist in this study. Atropine was administered prior to induction of anesthesia in 3 out of 6 BradyCI patients. In the remaining 3 BradyCI patient the anesthetist opted to accept the observed HR. No BradyCI patient required any more advanced interventions than giving a single dose of atropine.

Clonidine is also used outside the perioperative setting, both in adult and pediatric populations it is prescribed for several indications, including attention deficit hyperactivity disorder (ADHD), hypertension, muscle spasticity, opioid withdrawal, Tourette syndrome, headache, acute pain and nicotine dependence. There is an increase in usage in children with ADHD. The usage has doubled in the United States between 2003 and 2008 and had a FDA-approval (Food and Drug Administration) for ADHD in 2010. The increase of prescriptions and subsequent patients receiving clonidine treatment, has led to an increase in trends of unintentional clonidine medication exposures in the United States from 2000 to 2011, Figure 9.

**Figure 9** Trends for alpha-2 agonist medication exposures from 2000-2011. There was a significant trend over time for the overall increase of alpha-2 agonist calls to NPDS at 5% per year, CI 3.6-8.2. (P < .001). Clonidine by 3.3% per year.
Most common symptoms of patients accidentally exposed to clonidine presents with CNS (central nervous system) depression (e.g. drowsiness and lethargy) and bradycardia. Clonidine ingested in cases of accidental pediatric intoxication report an incidence of bradycardia between 9-14%. Very few patients require critical care interventions as intubation, ventilation, atropine and/or vassopressor support. The need for atropine or vasopressor administration during the PICU observation period was reported to be low (1.8 % and 0.4 %, respectively). Overall mortality is reassuringly very low. Clonidine has a reassuring safety profile but there is a risk that the incidence of accidental exposure will increase with the increase usage of clonidine in patients for ADHD and acute pain, together with the fact that the oral clonidine solution in Sweden is produced by the hospital pharmacy in a similar dark bottle as paracetamol, Picture 1.

### 6.4 Future perspectives

Premedication in children has been used since the 1940s with a variety of drugs and administration routes. The introduction of alpha-2 agonist into the field of pediatric anesthesia in the early 1990s has put forward alternatives to the previous considered golden standard of midazolam as premedication. Many studies and meta-analysis have been published reporting the efficacy and safety of Clonidine in producing preoperative sedation, reducing postoperative emergence delirium and reducing postoperative pain superior to benzodiazepines. The published data regarding the pharmacokinetics of clonidine as described above is limited due to limited population size and limited sampling. The European Medicines Agency and the European Union has highlighted the urgency for future trials to explore the efficacy, safety and pharmacokinetics of clonidine in different age groups of children. The upcoming CloSed trial will explore the efficacy, safety and pharmacokinetics of clonidine infusion compared to midazolam infusion for sedation in the pediatric intensive care setting. Blood samples for pharmacokinetic data will be collected with and analyzed with a minimal volume technique reducing the amount of blood drawn from each patient, which is essential in order to be able to include the small infants and neonates. The pharmacokinetic data will then be analyzed with population pharmacokinetics. These data will be from intravenous administration of clonidine.
Dexmedetomidine is widely used in Europe and Australasia as an oral premedication and the exploration of oral clonidine is also warranted.

Dexmedetomidine the new more selective alpha-2 agonist is widely used in the pediatric population. It is administered as an infusion in the pediatric intensive care setting but also for procedural sedation and premedication. There is an increasing interest in dexmedetomidine administered as a nasal aerosol which has similar beneficial properties as oral clonidine in the perioperative period, but has a more rapid onset of sedation when administered intranasally. Dexmedetomidine is seen as promising alternative to oral clonidine. Both these alpha-2 agonist has been not been associated with development of neurotoxicity or apoptosis, in fact in vivo experiments suggest that they may be protective when administered before exposure to routinely used anesthetic agents. Future trials are warranted in exploring the efficacy, safety and pharmacokinetics of intranasal administered dexmedetomidine as a premedication.
7  Conclusions

1.  **Study I**
Clearance is reduced in neonates and infants compared to adults and reaches 82% of the adult rate by 1 year of age. It is recommended to individualize and reduce the doses of clonidine in this age-group.

2.  **Study II**
The absorption of clonidine as nasal drops with our administration technique is low. Nasal administration of clonidine as nasal drops cannot be recommended due to great interindividual variability.

3.  **Study III**
Oral bioavailability of clonidine is reduced in children compared to adults and also compared to the complete bioavailability reported for both epidural and rectal clonidine administration of clonidine in children. Our results suggest that it would be necessary to administer at least twice the intravenous dose orally to get a similar effect of clonidine in children when compared with intravenous administration.

4.  **Study IV**
Clonidine administrated as a nasal aerosol does not improve the onset time of preoperative sedation even if using a dose as high as 7–8 mikrog·kg\(^1\). Nasal administration of clonidine as a nasal aerosol for preoperative sedation cannot be recommended if an onset time ≤ 30min is desired.

5.  **Study V**
The incidence of bradycardia following oral or intravenous premedication with clonidine at clinically relevant doses in a pediatric population is very low. Hence it does not appear rational to refrain from using clonidine as premedication in children only due to the potential risk for bradycardia.
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