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Article

How Do Circadian Rhythms Impact Spinal Anesthesia Effects and Surgical Stress Parameters?

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Abstract: Understanding how the time of day affects the pharmacokinetics and pharmacodynamics of anesthetic agents is essential for interpreting drug responses and variations in recovery from anesthesia. This study aimed to evaluate differences in the duration of spinal anesthesia and postoperative pain levels after cesarean delivery, focusing on the timing of anesthesia administration. Ninety women undergoing cesarean sections were divided into three groups based on the timing of intrathecal drug administration: group A (08:00-16:00), group B (16:00-00:00), and group C (00:00-08:00). Post-anesthetic evaluations assessed sensory and motor blockade and pain intensity at ten-minute intervals. Blood samples were collected before anesthesia and at 2, 4, 24, and 48 hours post-surgery to measure cortisol and C-reactive protein (CRP) levels. Results showed that group A patients experienced longer durations of motor and sensory blockade, delayed requests for postoperative analgesia, and lower pain scores (all $p < 0.05$). Significant differences in CRP levels were noted between group A and group B at 24 and 48 hours ($p < 0.05$).

Keywords: circadian rhythms; spinal anesthesia; local anesthetics; surgical stress

Introduction

Circadian physiology significantly impacts various physiological, pathophysiological, and pharmacological aspects, especially concerning anesthesia and surgery. This influence extends to several factors, from pharmacokinetics and pharmacodynamics of anesthetic agents, such as local anesthetics, to cardiovascular stress response, immunity and wound healing¹. Research indicates that circadian rhythms influence both the disposition and actions of various drugs, such as local anesthetics, and can affect therapeutic efficacy and toxicity based on dosing time², generating increased interest in the circadian variations of endogenous rhythms within anesthesia and surgical practices³. Consequently, the circadian clock may have critical clinical implications for surgical physiology, potentially resulting in varied responses to anesthesia and surgical interventions⁴. Nevertheless, the understanding of circadian rhythms in relation to neuraxial anesthesia remains limited. Our literature review reveals that numerous studies have emphasized the time-dependent efficacy of local anesthetics delivered via epidural and intrathecal routes⁵⁻¹³.

We previously acknowledged the potential influence of time-related factors on the effectiveness of intrathecal levobupivacaine, particularly concerning the timing of administration in relation to circadian rhythms, without accounting for individual variations in circadian organization as indicated by plasma cortisol levels (rhythm marker)¹⁵. To further investigate the time-dependent effects on spinal anesthesia, we designed a novel prospective study with stringent inclusion criteria. This approach ensured that all known confounding factors affecting circadian rhythms were controlled, and we confirmed the circadian organization of participants by measuring plasma cortisol levels before the induction of anesthesia. The study involved administering spinal anesthesia with a standard drug dosage at various times throughout the day to healthy women undergoing cesarean delivery.

Aim

The primary aim of this study was to evaluate potential temporal variations in the duration of spinal anesthesia and the postoperative pain experienced following cesarean delivery, with particular emphasis on the timing of anesthesia administration, including comparisons between daytime and nighttime. Furthermore, a secondary objective was to explore the possible temporal relationship between the timing of spinal anesthesia and the levels of C-reactive protein (CRP) and cortisol in the early postoperative period.

Methods

Participants received comprehensive verbal information regarding the study's objectives and the procedures for data and blood sample collection. Participation was entirely voluntary, allowing patients the option to withdraw at any point during the study. Additionally, written informed consent was secured from all individuals involved in the research.

The study included ninety primiparous to multiparous laboring women treated in our hospital for caesarean section under spinal anesthesia over different time periods. Participants were categorized into three groups based on the timing of intrathecal drug administration: group A (08:00-16:00), group B (16:00-00:00), and group C (00:00-08:00), with each group consisting of thirty subjects and an equal duration of eight hours. The chosen time frames for patient enrollment were coordinated with work shifts to facilitate the study's implementation. The research covered both scheduled and emergency caesarean sections under spinal anesthesia, with scheduled procedures assigned to group A and urgent cases allocated to groups B and C.

All participants participated in pre-surgery interviews to collect baseline data, which included demographic information, laboratory results, ASA physical status, and details about their feeding habits, sleep patterns, smoking and alcohol consumption, and levels of physical activity. All study participants reported maintaining a consistent lifestyle for at least five days leading up to their surgery. This included adherence to a regular light exposure schedule, a structured feeding and sleeping routine, and a consistent bedtime each night. They aimed for seven to eight hours of quality sleep, waking up around the same time daily, and following regular meal times. Additional inclusion criteria required participants to have an ASA physical status of I-II, a BMI below 30 kg/m², and singleton term pregnancies of more than 37 weeks gestation. Indications for cesarean deliveries included nonprogressive labor, cephalopelvic disproportion, abnormal cardiotocography, umbilical cord prolapse, placenta previa, repeat cesarean delivery, and maternal request. Exclusion criteria included shift work, abnormal coagulation profiles, psychoorganic syndromes, chronic pain syndromes, rheumatic diseases, and back pain.

The pre-anesthetic protocol involved establishing intravenous access through two 18G catheters and administering 500 ml of isotonic solution for prehydration, with no premedication provided. All participants received a single-shot spinal anesthesia, utilizing a standard dosage of 13 mg levobupivacaine (0.5%) combined with 0.02 mg fentanyl. The intrathecal injection was conducted at the L3–L4 or L4–L5 interspace using a 25-gauge Quincke needle, with a slow infusion rate exceeding

15 seconds while the patient was in a sitting position. Each participant underwent non-invasive monitoring of blood pressure, pulse oximetry, capnography, and electrocardiography, with vital signs documented throughout the procedure. Sensory and motor evaluations were conducted every minute until surgery commenced, and any side effects, including hypotension, bradycardia, nausea, vomiting, and shivering, were meticulously recorded. Post-anesthetic follow-up was conducted at 10-minute intervals to evaluate both sensory and motor blockade until full recovery, alongside assessing pain intensity at the first request for postoperative analgesia. Sensory blockade was measured using hot/cold tests and pin-prick stimulation, while motor blockade was assessed using the four-point modified Bromage scale, which ranges from 0 (full flexion of knees and feet) to 3 (complete inability to move legs or feet). Pain intensity was quantified using a numerical scale from 0 to 10, where 0 indicates no pain and 10 represents the worst pain imaginable. Blood samples of 3 ml were collected from each participant prior to anesthesia induction and at 2, 4, 24, and 48 hours post-surgery to analyze cortisol and CRP levels. Sampling occurred over a three-year period, spanning from December 2017 to February 2020. The protocol was exclusively implemented during the winter months (December to February), ensuring consistent temperature, humidity, and lighting conditions. Each participant was situated in a controlled environment, characterized by standard lighting and temperature, while being isolated from noise and electronic devices.

Statistical Analysis

Statistical analysis of the data was performed using the Statistical Package for the Social Sciences (SPSS), version 27.0 (IBM). The normality of quantitative variables was tested with Kolmogorov-Smirnov test. All quantitative parameters were expressed as mean values ± standard deviation (S.D.). For the statistical evaluation of the difference in the indicators between the three different groups analysis of variance was used (ANOVA). Multiple comparison was performed using the Least Significant Difference (LSD) test with corrected significance level at α = 0.05 according to Bonferroni’s correction. All statistical tests were two-sided and the results were considered statistically significant for p<0.05.

Results

The demographic characteristics of the patients were similar across the three groups, with no significant differences observed. No significant differences were observed among the three groups when comparing the time required for sensory block necessary for surgical procedures. The analysis included the duration of motor block until Bromage 0, the length of sensory block for both touch sensation and pin-prick, the interval from spinal anesthesia administration to the first request for postoperative analgesia, and the NRS scores at the time of the initial analgesic request, as detailed in Table 1.

Table 1. The duration of motor and sensory blockade, the time until the first request for postoperative analgesia, and the Numeric Rating Scale (NRS) score at the time of the initial analgesic request were evaluated based on the timing of spinal anesthesia administration, categorized into three groups: group A (08:00 - 16:00), group B (16:00 - 00:00), and group C (00:00 - 08:00).

Groups	A (n=30)	B (n=16)	C (n=16)	P value
Duration of motor blockade (min)	200.20±47.14	173.04±35.60@	135.47±31.59*	<0.05
Duration of sensory blockade (min)	269.60±44.26 #	219.10±26.20*	188.67 ±25.33	<0.05

Time to postoperative				
analgesic request (min)	243.40±50.70#	202.10±33.70	173.43±32.24	<0.05
NRS score at first analgesic request	4.34±0.72	4.53±0.67	5.47±0.85*	<0.05

Least Significant Difference test $\alpha = 0.01$. The significance level is $p < 0.05$. Statistically significant difference: @ compared to group A; *compared to groups A and B; #compared to groups B and C; ~ compared to group C.

ANOVA revealed statistically significant intergroup differences in the duration of motor and sensory blockade, time of spinal anesthesia’s administration to first postoperative analgesic request and NRS scores (all $p < 0.05$). In particular, in post-hoc analysis the following statistically significant differences were observed: in the duration of motor blockade in group C versus groups A (mean difference $\pm$ standard error, -64.73 ± 15.55 min, $p < 0.001$) and B (-37.57 ± 4.01 min, $p = 0.005$) and in group B versus group A (-27.16 ± 11.54 min, $p = 0.02$); in the duration of sensory blockade in group C versus groups A (-80.93 ± 18.93 min, $p < 0.001$) and B (-30.43 ± 0.87 min, $p = 0.027$) and in group B versus group A (-50.5 ± 18.06 min, $p < 0.001$); in the duration of time of anesthesia administration to first postoperative analgesic request in group A versus groups B (41.3 ± 17.0 min, $p = 0.006$) and C (69.97 ± 18.46 min, $p < 0.001$); in NRS scores at first analgesic request in group C versus groups A and B (all $p < 0.001$).

Group A exhibited the longest duration of both motor and sensory blockade when compared to groups B and C ($p < 0.05$), while group C demonstrated the shortest duration relative to groups A and B ($p < 0.05$). Additionally, the interval from the administration of anesthesia to the first request for postoperative analgesia was significantly longer in group A than in groups B and C ($p < 0.05$). No significant difference was observed in this interval between groups B and C ($p = 0.055$). Furthermore, group C recorded higher Numerical Rating Scale (NRS) scores at the time of the first analgesic request compared to groups A and B (all $p < 0.001$), with no notable difference in NRS scores between groups A and B ($p = 0.459$).

All groups were assessed for serum cortisol levels before and after surgery at 2, 4, 24, and 48 hours post-operation. ANOVA revealed no statistically significant differences in preoperative or postoperative cortisol levels at these time points ($p = 0.692$), with all p -values exceeding 0.05, as shown in Table 2.

Table 2. Preoperative and postoperative serum cortisol levels at 2h, 4h, 24h and 48h after the operation.

Cortisol levels (nmol/L)	Group A	Group B	Group C	p- value
preoperative levels	398.65 $\pm$ 217.25	445.82 $\pm$ 356.45	446.17 $\pm$ 241.45	0.750
2 h postoperative	396.07 $\pm$ 235.45	419.51 $\pm$ 304.88	430.55 $\pm$ 202.59	0.863
4 h postoperative	356.45 $\pm$ 184.67	375.45 $\pm$ 236.60	479.13 $\pm$ 258.25	0.088
24 h postoperative	450.16 $\pm$ 246.76	474.44 $\pm$ 312.80	434.98 $\pm$ 212.48	0.840
48 h postoperative	398.59 $\pm$ 162.85	400.26 $\pm$ 185.10	335.71 $\pm$ 156.13	0.245

Least Significant Difference test $\alpha = 0.01$. The significance level is $p < 0.05$.

All groups were assessed for serum CRP levels before and after the operation at 2, 4, 24, and 48 hours post-surgery, as shown in Table 3.

Table 3. Preoperative and postoperative serum cortisol levels at 2h, 4h, 24h and 48h after the operation.

Cortisol levels (nmol/L)	Group A	Group B	Group C	p- value
preoperative levels	398.65 ± 217.25	445.82 ±356.45	446.17 ±241.45	0.750
2 h postoperative	396.07± 235.45	419.51 ±304.88	430.55 ±202.59	0.863
4 h postoperative	356.45 ±184.67	375.45± 236.60	479.13±258.25	0.088
24 h postoperative	450.16 ±246.76	474.44 ±312.80	434.98 ±212.48	0.840
48 h postoperative	398.59 ±162.85	400.26 ±185.10	335.71 ±156.13	0.245

Least Significant Difference test $\alpha = 0.01$. The significance level is $p < 0.05$.

ANOVA indicated that there were statistically significant differences in postoperative CRP levels among the groups at both 24 hours ($p=0.039$) and 48 hours ($p=0.028$), with all p-values being less than 0.05. Mean CRP concentrations rose across all groups at both 24 and 48 hours following surgery. Post-hoc analysis identified significant differences in CRP levels between group B and group A at 24 hours ($p<0.05$) and 48 hours ($p<0.05$), as well as between group B and group C at 48 hours ($p<0.05$).

Discussion

In this study, we found that patients who underwent spinal anesthesia for cesarean sections between 08:00 and 16:00 (group A) experienced a longer duration of both motor and sensory blockade, a delayed first request for postoperative analgesia, and lower pain scores (all $p<0.05$). Conversely, we observed a reduced duration of both motor and sensory blockade in patients treated between 00:00 and 08:00 (group C) (all $p<0.05$). Additionally, our findings indicated that the request for postoperative analgesia was later between 08:00 and 00:00 (groups A and B) and occurred earlier between 00:00 and 08:00 (group C) (all $p<0.05$). Regarding the Numeric Rating Scale (NRS) scores at the time of the first analgesic request, we noted that pain intensity was significantly higher at night (group C) compared to other time intervals ($p<0.05$).

Our research supports previous studies that demonstrate circadian variations in anesthetic effectiveness. Costa-Martins et al⁷. and Debon et al⁸., found that the effects of epidural anesthesia lasted longer during the day than at night. Vieira WS et al. identified a diurnal pattern in spinal analgesia during labor, with peaks at 00:00 and 12:00 hours, while labor analgesia peaked between 02:00 and 05:59 hours⁹. Moataz Morad El-Tawil et al., noted a shorter duration of intrathecal bupivacaine in the evening and nighttime¹⁰. Chassard et al. reported a 25% variation in the duration of intrathecal plain bupivacaine during daytime, peaking around noon¹¹. Kılıçarslan et al. found that patients undergoing inguinal hernia and anorectal surgeries under spinal anesthesia in the morning (06:00–12:00) had a longer post-operative first analgesic requirement time¹². Lee et al. showed that the time to the first postoperative analgesic and the recovery of S1 sensation to pinprick were significantly longer in the noon group (12:00 to 16:00)¹³. However, unlike our findings, these authors did not find significant differences in sensory and motor block duration. In our previous study, we reported the shortest spinal block duration at night, the longest during the day, and peak analgesia

duration at noon¹⁴. In contrast, Scavone et al¹⁵. and Shafer et al¹⁶., concluded that the time of day did not significantly influence the duration of analgesia from intrathecal local anesthetics or opioids.

The pharmacokinetics and pharmacodynamics of local anesthetics are significantly influenced by molecular clocks^{17,18}. The timing of drug administration can be crucial in determining essential pharmacokinetic parameters such as overall exposure, bioavailability, clearance, peak concentration, and half-life. Research indicates that variations between day and night, particularly the nocturnal lows in blood pressure, cardiac output, stroke volume, and hepatic blood flow, may lead to differences in drug distribution and metabolism¹⁹⁻²³. Circadian fluctuations in plasma protein binding, the penetration of local anesthetics into erythrocytes, tissue drug penetration, and cytochrome P450 activity have been observed^{2,24-25}. Notably, protein binding tends to increase during nighttime, while metabolic processes are more pronounced during the active daytime hours, which may account for the previously noted temporal variations in drug kinetics. Additionally, various ion channels, transporters, and efflux pumps show circadian variations in tissue expression, which can potentially impact the transport and efflux of drug substrates in a tissue-specific manner². Conversely, for numerous drugs, significant differences in their effects can arise from varying dosing times, even when drug exposure remains constant, suggesting the involvement of alternative pharmacodynamic mechanisms. In clinical practice, numerous factors can influence the pharmacokinetics and pharmacodynamics of anesthetic agents, potentially obscuring or eliminating circadian variations. As a result, there is considerable inter-individual variability in the pharmacokinetic and pharmacodynamic parameters of anesthetics²⁶.

Our study revealed variations in Numerical Rating Scale (NRS) scores between day and night, indicating that circadian rhythms affect pain sensitivity. Inès Daguet and colleagues support this notion, positing that pain sensitivity is controlled by a strong circadian component and a modest homeostatic sleep-related component²⁷. An experimental study confirmed lower pain sensitivity during daytime and increased sensitivity at night²⁸, consistent with previous meta-analyses suggesting maximum pain sensitivity occurs peak pain sensitivity is in the middle of the night²⁹. Furthermore, animal studies indicate downregulation of opioid receptors during morning, early afternoon, and late evening hours³⁰. Cicekci et al. found that pediatric patients undergoing surgery from 01:01 to 07:00 experienced more pain compared to patients who underwent surgery at other times of the day³¹. Research by Pan et al³². and Aya et al³³. showed that morning VAPS scores were lower during neuraxial labor analgesia assessment. Desai et al. suggested that requests for neuraxial analgesia and labor onset occurring in the evening and night resulted in higher pain scores compared to morning and afternoon instances³⁴. Additionally, Arslan, Gülten et al. reported that patients who underwent surgery in the morning (08:00–12:00) experienced less postoperative pain and required less analgesia than those operated on in the afternoon (12:00–16:00)³⁵. Costa-Martins et al⁷. and Deng Jiali et al³⁶. noted increased pain scores in patients with epidural analgesia during nighttime labor. However, Debon et al. found no significant differences in Visual Analog Scale pain scores throughout the day⁸.

In our study, we found that women who received spinal anesthesia at night reported higher pain scores compared to those treated at other times. This may be influenced by factors such as the urgency of the surgery, pre-existing pain, and anxiety. Additionally, sleep deprivation could affect pain perception by impairing opioid receptor function in the mesolimbic circuit, particularly the μ and δ receptors, which reduces endogenous opioid levels and central receptor sensitivity. This can lead to diminished analgesic effectiveness and heightened pain perception^{29,37-38}. Evidence also supports hyperalgesia in healthy individuals following substantial perturbations of sleep³⁹. Moreover, the nocturnal decline in pain-regulating hormones like melatonin, corticosterone, progesterone, and endogenous opioids may contribute to the observed variations in pain perception, as these hormones decrease the pain perception threshold⁴⁰. Cortisol levels typically peak in the morning, while melatonin acts at night, with research indicating reduced analgesic needs during these periods⁴⁰. Lastly, obstetric factors, such as the nature of labor onset and parity, may also influence postoperative pain severity^{33,41}.

Postoperative recovery has been assessed through the monitoring of blood biomarkers, specifically cortisol and C-reactive protein (CRP). CRP, an acute phase reactant, increases after surgery due to inflammation, tissue damage, or blood loss⁴². However, CRP response in individual patients is highly variable⁴³, and in some patients may be incomplete or even absent⁴². Cortisol levels also rise due to the physiological stress of surgery⁴⁴. Research indicates that wide-ranging neuraxial analgesia with local anesthetics can reduce maternal stress during cesarean sections⁴⁵⁻⁴⁶.

Our study aimed to explore the postoperative levels of CRP and cortisol in relation to the timing of anesthesia administration. We found that CRP levels rose following surgery in all groups, peaking around 48 hours post-operation. Notably, women who had surgery between 16:00 and 00:00 (group B) showed significantly higher CRP levels at 24 and 48 hours ($p < 0.05$). Literature suggests that CRP is affected by circadian rhythms, with some studies supporting our findings. For example, Rudnicka et al. noted diurnal variations in serum CRP, peaking at 3 PM, with higher morning levels compared to evening⁴⁷. Wetterö et al⁴⁸. and Izawa et al⁴⁹. also reported increased salivary CRP and total protein levels at awaking time in the morning versus evening. In contrast, Meier-Ewert et al⁵⁰. found no significant diurnal variation in serum levels throughout the day, and Mills et al⁵¹. found no significant changes in Non-apneics over 24 hours. However, the study's author noted diurnal variability in C-reactive protein (CRP) levels in individuals with obstructive sleep apnea, with higher levels during the day than at night. It is crucial to note that previous studies focused on the general population rather than surgical patients. Only one study has investigated circadian rhythms in postoperative CRP levels after orthopedic surgery, which did not find significant differences in plasma CRP concentrations 24 hours post-surgery⁵². The limited research on circadian effects on postoperative CRP levels in surgical patients highlights the need for further studies in this area.

Strengths and Limitations

To the best of our knowledge, the impact of circadian rhythms on C-reactive protein (CRP) levels following cesarean sections performed under spinal anesthesia has not been previously explored, which represents a notable strength of our research. However, our study does have certain limitations. We did not measure CRP levels beyond 48 hours, preventing us from tracking the progression of CRP values until patient discharge. Additionally, we did not assess pre-existing pain in women who underwent emergency cesarean sections, a variable that could potentially affect pain outcomes. Furthermore, while we ensured that study participants were placed in a standard room free from noise and electronic devices, we could not regulate interactions with visitors, electronic communications (such as smartphones), or hospital routines. These elements may influence diurnal rhythms. Consequently, the applicability of our findings may be limited, as the study was specifically conducted in the context of cesarean sections.

Conclusions

Our study provides evidence that circadian rhythms, especially concerning the timing of anesthesia administration, play a crucial role in determining the efficacy of spinal anesthesia and the severity of postoperative pain in both elective and emergency cesarean deliveries, as well as in serum CRP levels after surgery. We assume that the observed variations in CRP levels across different time intervals may reflect the impact of circadian rhythms on spinal anesthesia. Recognizing these variations in the analysis of laboratory results could enhance postoperative monitoring, particularly in light of the current trend towards faster patient discharges. Our findings possess clinical relevance, particularly in light of the increasing prevalence of elective surgeries scheduled for the evening. Future applications of circadian rhythm insights could facilitate the optimal timing of medical procedures and the administration of anesthetics or analgesics, thereby enhancing patient care. Future research on circadian principles could optimize medical interventions and improve patient outcomes, necessitating further investigation into individual chronotypes.

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Informed Consent Statement: Informed consent statement was obtained from all subjects involved in the study. Written informed consent has been obtained from the patients to publish this paper.

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