

## Compare the Efficacy of Intravenous Magnesium Sulphate and Dexmedetomidine in Reducing the Hemodynamic Stress Response during Laryngoscopy and Endotracheal Intubation

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### Abstract

**Objective:** This study aimed to compare the efficacy of intravenous Magnesium Sulphate and Dexmedetomidine in reducing the hemodynamic stress response during laryngoscopy and endotracheal intubation.

**Methods:** The present study included a total of 60 patients of either sex aged between 18 to 60 years who underwent surgery under general anaesthesia. The patients were randomly allocated into two groups, Group M (Magnesium Sulphate) and Group D (Dexmedetomidine) of 30 patients each. Group M patients were administered intravenous 25 mg/kg of 50 % Magnesium Sulphate in normal saline (10ml total) and Group D patients were received intravenous Dexmedetomidine 1 mcg/kg in normal saline (10ml total), 10 minutes before intubation. In the present study demographic variables, anthropometric variables, ASA physical status and duration of surgery was comparable between two groups.

**Results:** Mean heart rate, mean systolic blood pressure, mean diastolic blood pressure, mean MAP in Magnesium Sulphate (M) group was significantly higher as compared to that of Dexmedetomidine (D) group throughout the study period except at baseline. Hence Dexmedetomidine (D) group is better than Magnesium Sulphate (M) group Both the drugs had minimal side effects with no significant difference between the groups. Overall, Dexmedetomidine provided a better Hemodynamic study and was found to be superior when compared to Magnesium sulphate.

**Conclusion:** The sympathetic reactions to laryngoscopy and intubation have been suppressed using a variety of medication combinations with varying degrees of efficacy. Overall, Dexmedetomidine provided a better Hemodynamic study and was found to be superior when compared to Magnesium sulphate.

**Keywords:** Laryngoscopy, Tracheal Intubation, Mean Arterial Pressure (MAP), Hemodynamic Abnormalities, Tachycardia, Hypertension, Magnesium Sulphate, Dexmedetomidine (DEX), Fentanyl.

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### Introduction

Laryngoscopy and tracheal intubation are known to cause a wide range of

hemodynamic abnormalities, including tachycardia, hypertension, and arrhythmias

of the heart, since they release catecholamines and trigger sympathetic reactions. Using a laryngoscope, inserting a tracheal tube, and inflating a cuff all contribute to stimulation of the supra glottic region, which is thought to be the primary cause of sympathoadrenal reactions. Reflex alterations in the cardiovascular system following laryngoscopy and intubation typically increase blood pressure by 40-50% and heart rate by 20% if no special steps are taken to prevent hemodynamic reaction. Alterations in hemodynamics are often temporary. People who are healthy might be able to tolerate it just well, but those who have pre-existing conditions like hypertension, coronary artery disease, cerebrovascular disease, myocardial infarction, or thyrotoxicosis might find it harmful. [1-5]

Multiple medications, including opioids, calcium channel blockers, beta blockers, alpha-2 agonists, Magnesium Sulphate, local anaesthetics etc., have been utilized to dampen these reactions. Dexmedetomidine (DEX), a short-acting, highly selective alpha2-adrenoreceptor agonist, is used for its sedative, analgesic, and anxiolytic effects without causing any respiratory depression. Anxiety and uneasiness might be calmed with this drug just before general anaesthesia is administered. It has been well-documented that the laryngeal stress response can be adequately mitigated by administering DEX intravenously (IV) prior to surgery. However, it could lead to potentially dangerous hemodynamic side effects like low blood pressure and a sluggish heart rate. Recovery times are also known to lengthen when IV DEX is used, as it has a sedative effect. Magnesium sulphate is a CNS depressant that works by inhibiting both dopamine and noradrenaline release from the sympathetic nervous system. The vasodilation that occurs as a result of the reduced release of catecholamines is a side effect. Dexmedetomidine, a highly selective 2 adreno receptor agonist, counteracts the hemodynamic fluctuation that occurs at the

time of extubation due to enhanced sympathetic activation by decreasing the sympathetic outflow and noradrenergic activity. It is possible that elevated serum magnesium levels also prevent the release of catecholamines. [6-9]

This study aimed to compare the efficacy of intravenous Magnesium Sulphate and Dexmedetomidine in reducing the hemodynamic stress response during laryngoscopy and endotracheal intubation.

### **Materials and Methods**

**Study Design:** Prospective, controlled, randomized & double blinded study.

**Place of Study:** The study was carried out in the Department of Anaesthesiology & critical care. F.H. Medical College & Hospital, Etmadpur Agra

**Study Group:** The prospective randomized & double blinded study was conducted on 60 patients in the age group of 18-60 years divided into two groups (30 each) after obtaining approval from the Institutional Ethical Committee. Informed, written consent from all the patients were obtained from eligible patients. The patients were visited a day prior to surgery for pre-anaesthetic review and standard institutional pre-operative advice was given.

### **Inclusion criteria:**

- Adult Patients from either sex aged between 18 to 60 years who underwent surgery under general anaesthesia.
- ASA physical status I and II.
- Scheduled for elective surgery under general anaesthesia.

### **Exclusion criteria:**

- Lack of patient's consent.
- ASA physical status > II.
- Patients with anticipated difficult intubation:- Mallampati grade III & IV.
- BMI >35, (Morbid obesity).
- The sign of cardiovascular, respiratory diseases.
- Hepatic dysfunction.

- History of drug abuse.
- History of drugs like Calcium channel blockers, narcotics, hypnotics, beta blockers.
- Patients with known hypersensitivity to Magnesium sulphate and Dexmedetomidine.

**Methodology:** The study was initiated after obtaining approval from the institutional ethical committee and children were enrolled after obtaining informed consent from the patients side. This prospective, controlled, randomized & double blinded study was carried out in the Department of Anaesthesiology & Critical Care, F.H. Medical College & Hospital, Etmadpur Agra. Subjects in the age group of 18-60 years, who underwent elective surgery under general anaesthesia as a part of their therapy and classified as American society of anaesthesiologists (ASA) I and ASA II were included. All enrolled patients underwent pre-anaesthesia evaluation, one day prior to the surgery. On arrival in operation theatre Baseline parameters SpO<sub>2</sub>, pulse rate, Systolic blood pressure, Diastolic blood pressure and mean arterial blood pressure was recorded. IV lines with 18 G cannula was secured and all the patients were pre-medicated with injection glycopyrrolate 10 mcg/kg, fentanyl 2mcg/kg, Injection Ondansetron 0.1 mg/kg. Simultaneously they were pre oxygenated with 100% oxygen for 3 minutes. The patients were randomly allocated into two groups, Group M (Magnesium Sulphate) and Group D (Dexmedetomidine) of 30 patients each.

Group M patients were administered intravenous 25 mg/kg of 50% magnesium sulphate in normal saline (10ml total) and Group D patients were received intravenous dexmedetomidine 1 mcg/kg in normal saline (10ml total), 10 minutes before intubation. Patients were induced with IV propofol 1.5 mg/kg followed by IV succinylcholine 1.5 mg/kg. Hemodynamic measurements (HR/SBP/DBP/MAP) were recorded, before induction (baseline), after induction, at the time of laryngoscopy and endotracheal intubation and then at 1, 3, 5, 10, minutes after intubation.

Intubation was performed with cuffed endotracheal tube of appropriate size after direct laryngoscopy. Anaesthesia was maintained with 50%O<sub>2</sub>+50%N<sub>2</sub>O delivered through bain's circuit using IPPV and propofol infusion (50 mcg/kg/min). Muscle relaxation was achieved with injection Vecuronium 0.1mg/kg followed by incremental doses of Vecuronium (0.02mg/kg).

At the end of surgery patients were reversed with neostigmine 0.05mg/kg and injection glycopyrrolate (0.01 mg/kg) after onset of spontaneous respiration. Any prevalence of laryngospasm, bronchospasm, desaturation or any intra-operative complications were recorded and managed according to standard protocols. Patients were shifted to recovery room any immediate postoperative complications were noted.

### Observation Chart

**Table 1: Age Distribution**

Age Group	Group M (n=30)		Group D (n=30)	
	Number of Patients	Percentage	Number of Patients	Percentage
18-30 years	7	23.3	8	26.7
31-40 years	10	33.3	9	30.0
41-50 years	6	20.0	7	23.3
51-60 years	7	23.3	6	20.0
<b>Total</b>	30	100.0	30	100.0
<b>Mean Age</b>	39.37 ±10.89		39.13 ±11.46	
<b>Statistical Inference</b>	<b>p value: 0.578</b>			

**Table 2: Sex Distribution**

Sex	Group M (n=30)		Group D (n=30)	
	Number of Patients	Percentage	Number of Patients	Percentage
Male	17	56.7	18	60.0
Female	13	43.3	12	40.0
Total	30	100.0	30	100.0
Statistical Inference	Chi- Square- 0.06857 P Value-0.793			

**Table 3: ASA Physical Status**

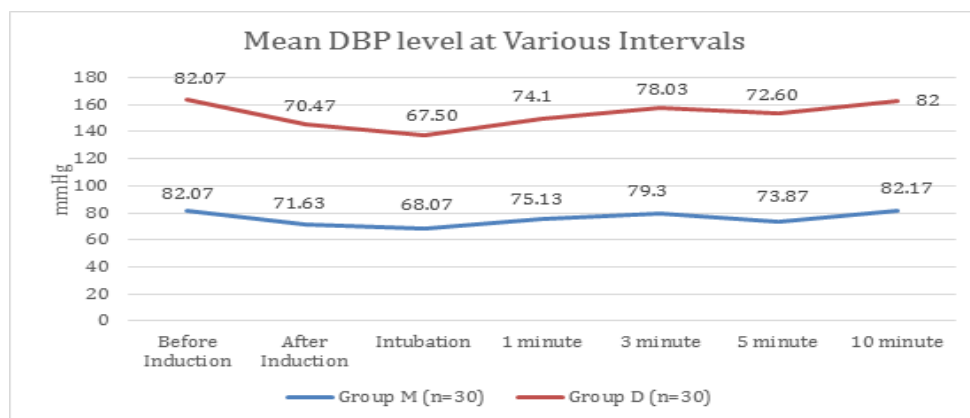
ASA Physical Status	Group M (n=30)		Group D (n=30)	
	Number of Patients	Percentage	Number of Patients	Percentage
ASA-I	22	73.3	24	80.0
ASA-II	8	26.7	6	20.0
Total	30	100.0	30	100.0
Statistical Inference	Chi- Square- 0.37267 P Value-0.541			

**Table 4: Anthropometric Variables**

Anthropometric Variables	Group M (n=30)		Group D (n=30)		p value
	Mean	±SD	Mean	±SD	
Height (cm)	158.60	±5.57	160.40	±5.97	0.537
Weight (kg)	69.80	±7.31	71.90	±5.74	0.247
BMI (kg/m <sup>2</sup> )	27.84	±3.86	28.06	±3.13	0.216

**Table 5: Duration of Surgery**

Duration of Surgery	Group M (n=30)		Group D (n=30)	
	Mean	±SD	Mean	±SD
Duration of Surgery (minutes)	91.00	±9.00	91.43	±9.57
Statistical Inference	p value: 0.556			



**Figure 1: Mean DBP level at Various Intervals**

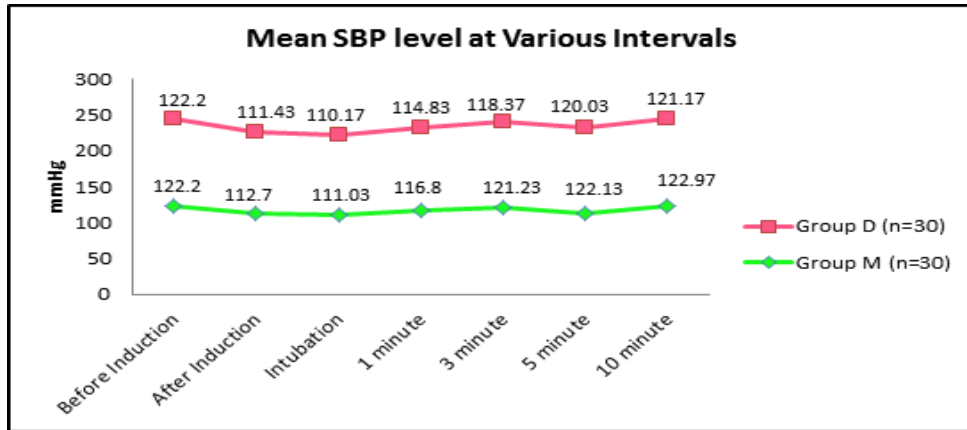


Figure 2: Mean SBP level at Various Intervals

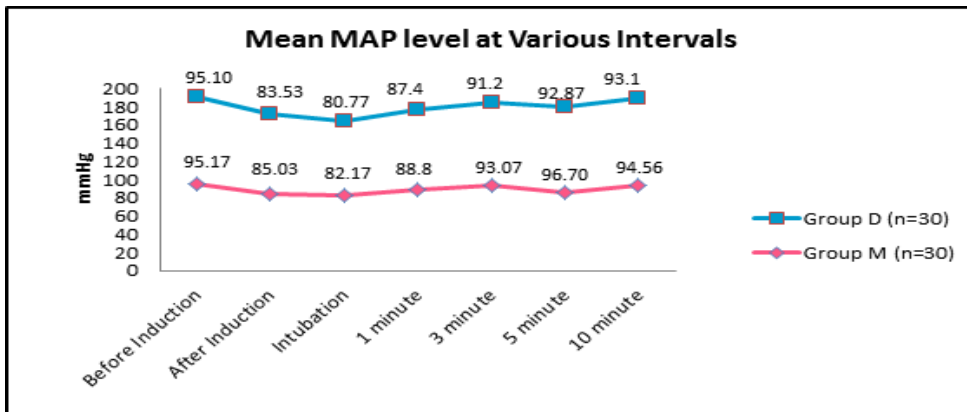


Figure 3: Mean MAP level at Various Intervals

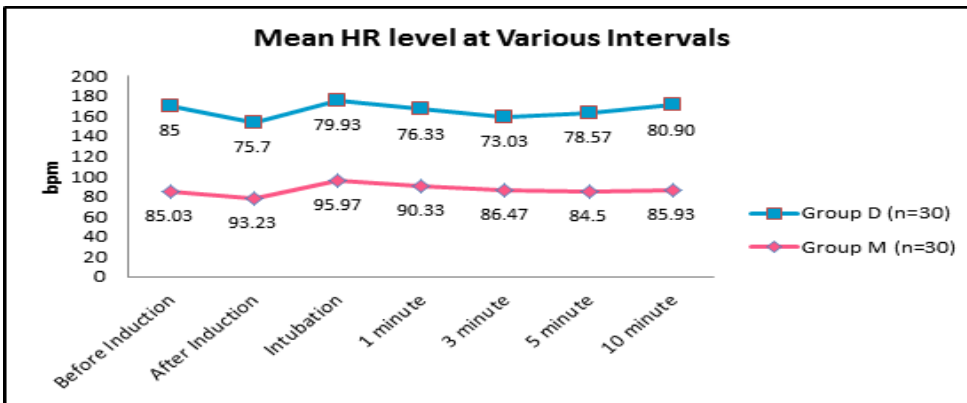


Figure 4: Mean HR level at Various Intervals

Table 6: Side Effects

ASA Physical Status	Group M (n=30)		Group D (n=30)	
	Number of Patients	Percentage	Frequency	Percentage
Bradycardia	1	3.3	2	6.7
Hypotension	3	10.0	2	6.7
Bradycardia with hypotension	1	3.3	1	3.3
No adverse Effects	25	83.3	25	83.3
<b>Total</b>	30	100.0	30	100.0
Statistical Inference	Chi- Square- 0.53333 P Value-0.911			

## Results

1. Mean heart rate in Magnesium Sulphate (M) group was significantly higher as compared to that of Dexmedetomidine (D) group throughout the study period except at baseline. Hence Dexmedetomidine (D) group is better than Magnesium Sulphate (M) group with respect to mean heart rate.
2. Mean systolic blood pressure in Magnesium Sulphate (M) group was significantly higher as compared to that of Dexmedetomidine (D) group throughout the study period except at baseline. Hence Dexmedetomidine (D) group is better than Magnesium Sulphate (M) group with respect to mean systolic blood pressure.
3. Mean diastolic blood pressure in Magnesium Sulphate (M) group was significantly higher as compared to that of Dexmedetomidine (D) group Throughout the study period except baseline. Hence Dexmedetomidine (D) group is better than Magnesium Sulphate (M) group with respect to mean diastolic blood pressure.
4. Mean MAP in Magnesium Sulphate (M) group was significantly higher as compared to that Dexmedetomidine (D) group throughout the study period except at baseline. Hence Dexmedetomidine (D) group is better than Magnesium Sulphate (M) group with respect to mean mean arterial blood pressure.
5. Hypotension was the most common adverse events in the present study included 3 patients in Group M and 2 patients in Group D. The other adverse events observed in the present study were bradycardia, and bradycardia with hypotension. Both the groups were comparable in terms of incidence of adverse events (p value = 0.911). Both the drugs had minimal side

effects with no significant difference between the groups.

Overall, Dexmedetomidine provided a better Hemodynamic study and was found to be superior when compared to Magnesium sulphate.

### Statistical Analysis:

Data was checked for accuracy and completeness then coded and entered into (Statistical Package for the Social Sciences) version 23.0 for analysis. The results presented in frequency tables, cross tabulations and figures. Categorical data are presented as frequency with percentages. Continuous data with normal distribution are presented as mean with standard deviation. Independent sample t-test was used to evaluate the difference between groups when data was normally distributed and Chi-square test was employed when the variables were not normally distributed. A p value <0.05 was considered statistically significant.

### Discussion

Endotracheal intubation is an essential component of general anaesthesia. It serves in the maintenance of patency of upper airway, proper ventilation, reduction in the risk of aspiration, and delivery of the inhalational anaesthetic agents to the patients through breathing circuits. Laryngoscopy and tracheal intubation are thought to be the most critical incidence during induction of general anaesthesia which stimulate somatic and visceral nociceptive afferents fibers which induce reflex sympato-adrenal responses associated with enhanced neuronal activity in the cervical sympathetic efferent fibers. Sympathetic stimulation from laryngoscopy and endotracheal intubation causes a significant increase in the plasma concentration of catecholamines (adrenaline and noradrenaline) that can provoke left ventricular failure, renal failure, surgical bleeding, cerebral hemorrhage and myocardial ischemia in anaesthetized patients. The mechanism of

this may be that, vasoconstriction, increased myocardial work, a demand for increased coronary flow, narrowed coronary arteries cannot accommodate the increased flow, and parts of the myocardium may receive insufficient oxygen. [9-14]

Laryngoscopy and intubation produce a reflex sympathetic discharge which causes adverse hemodynamic changes such rise in the arterial blood pressure, heart rate, pulmonary arterial pressure and wedge capillary pressure and can even cause cardiac arrhythmias additionally these laparoscopic surgeries also incur an overall stress because of the pneumoperitoneum caused by carbon dioxide insufflation. These changes in blood pressure and HR are maximum in the first 1 minute and last for 10 minutes after the procedure. To attenuate these deleterious effects various methods and drugs have been tried Nowadays there is a vogue of opioid free anaesthesia where multiple drugs like dexamethasone, lignocaine, magnesium sulphate, paracetamol, dexmedetomidine are being used intraoperatively. Anaesthesiologists are in constant search of a multimodal approach to blunt these unwanted responses. [15-22]

Common factors precipitating the pressor response to laryngoscopy and intubation are light planes of anaesthesia, prolonged time for the procedure, elevation of vagally innervated posterior part of epiglottis by straight/Miller blade, anatomically difficult view, greater force used to displace the tongue and more manipulations/attempts at laryngoscopy and intubation. Several drugs and maneuvers have been used for mitigating this stress response with variable benefits and side effects. [23]

Dexmedetomidine is a  $\alpha_2$  receptor selective and specific adrenergic agonist. These drugs by virtue of their sympatholytic (i.e. antihypertensive and negative chronotropic) action, attenuate the hemodynamic response following laryngoscopy and endotracheal intubation.

It has sedative, analgesic as well as sympatholytic properties. It has also been shown to be effective in maintaining hemodynamic stability during intubation and extubation without prolonging recovery. It also helps in attenuating airway reflex response to tracheal extubation. The unique property of dexmedetomidine is that it produces minimal respiratory depression so FDA has approved it for ICU sedation for less than 24 hours. Moreover, Dexmedetomidine also reduces the requirements of other anaesthetic agents like volatile anaesthetics and thiopentone sodium. [23-27]

Magnesium is well known to block the release of catecholamines from both adrenergic nerve terminals and the adrenal gland, and intravenous magnesium sulfate inhibits catecholamine release associated with laryngoscopy. Moreover, magnesium produces vasodilator effect by acting directly on blood vessels, and high-dose magnesium. Magnesium sulfate is a cerebral depressant which act by blocking NMDA receptor in CNS and by decreasing sympathetic outflow. It has a calcium antagonist property by competing with Ca for different channels in membrane and so can modify Ca mediated responses. Because of its NMDA receptor blocking action magnesium has been widely used for acute, chronic and postoperative pain management not as primary but as secondary analgesic. Because magnesium can decrease release of catecholamine from adrenal medulla and adrenergic nerves it can be safely use to suppress the effect of laryngoscopy and to provide cardiovascular stability. [28-30]

Therefore the present study was carried out to evaluate the efficacy of intravenous magnesium sulphate and dexmedetomidine in suppressing hemodynamic response to laryngoscopy and endotracheal intubation in adult patients undergoing surgery under general anaesthesia. Similar findings were observed in the previous studies done by Krishna Chaithanya et al, Bidyut Borah et

al, Mahajan L et al, Pathak et al, Vivek Arora et al, Ghodki P et al and Prithiv Rishardhan et al. Previous studies also reported that the mean age, weight, height and duration of surgery of both the groups were comparable. There was no significant difference amongst the groups with regard to demographic and clinical characteristics. Regarding the efficacy of intravenous magnesium sulphate and dexmedetomidine in suppressing hemodynamic response to laryngoscopy and endotracheal intubation we observed that Mean changes in DBP, SBP, MAP and HR in group D after induction, at intubation, at 1 minute, 3 minutes, 5 minutes and 10 minutes after intubation was statistically significant ( $p$  value =  $<0.05$ ) compared to group M. This further emphasizes the impact of Dexmedetomidine in stabilizing the hemodynamics of patient during and after laryngoscopy and intubation. [31-34]

In our study we found that with respect to heart rate a significant decrease in heart rate was observed in Dexmedetomidine group between baseline and after induction interval followed by a slight increase at intubation and then 1 minute, 5 minutes after intubation intervals. Statistically, the change from baseline values was significant at all the times intervals. In contrast, Magnesium sulphate group, continuous increase in heart rate was observed till 3 minutes post intubation interval, followed by decrease between 5 and 10 minutes post intubation intervals. As compared to baseline in Magnesium sulphate group, mean heart rate was significantly higher at all time intervals.

Comparison of mean heart rate between two groups showed a significantly higher mean value in Magnesium sulphate group as compared to Dexmedetomidine group at all the follow up intervals except at baseline. With respect to blood pressure (SBP, DBP, and MAP) too, in both the groups a significant decrease in mean values was observed after induction followed by a significant increase 1 minute

after intubation and then a gradual decrease till 5 minutes after intubation. On comparing the two groups, at all the follow up intervals, mean blood pressure was found to be significantly lower in Dexmedetomidine group as compared to Magnesium sulphate group.

The purpose of pre-operative medications is to control the hemodynamic stress induced by intubation. This is done generally by using such agents that bring down the hemodynamic parameters following stress could be manageable. This is done by inducing the receptors in body to be minimally affected by the stress generated by intubation. There have been several recent reports of improved postoperative pain control from the addition of magnesium to epidural or spinal infusions during surgery. Magnesium has no primary analgesic activity but may have value as a secondary analgesic, enhancing the actions of more established pain medication. Similar findings were also found on several studies which corroborate the finding of the hemodynamic attenuation by the drugs.

Krishna Chaithanya et al in their study compared the effectiveness of intravenous Magnesium sulphate and Dexmedetomidine in suppressing the cardiovascular stress response. Similar to the present study their observation was both magnesium sulphate and dexmedetomidine attenuated the rise in systolic and diastolic blood pressure, however in their study magnesium failed to attenuate increase in the heart rate which is less than 10 beats/ min. compared to dexmedetomidine which effectively controlled the rise heart rate following intubation. [35]

Bidyut Borah et al in their study the effectiveness of dexmedetomidine (D), clonidine (C) and magnesium sulfate (M) in attenuating the hemodynamic response to laryngoscopy in patients undergoing surgeries under general anaesthesia. In their study their observation regarding the above was that comparison of three drugs reveals



that hemodynamic variables were similar within the groups at baseline and at 5 minutes before intubation while there were significant difference between them starting from just after the intubation, 30 sec after intubation then 1 minute, 2 minutes, 3 minutes, 4 minutes, 5 minutes, 10 minutes after intubation and Dexmedetomidine (1mcg/kg) in comparison to magnesium sulfate (30 mg/kg) and clonidine (1 mcg/kg) is far more effective in blunting the hemodynamic response to laryngoscopy in their study. [36]

Mahajan L et al in their study determined the attenuation of the pressor responses to laryngoscopy and endotracheal intubation with intravenous dexmedetomidine versus magnesium sulphate under bispectral index -controlled anaesthesia. Their observation was that SBP, DBP and HR fell in the Dexmedetomidine and Magnesium Sulphate groups. There was no significant changes in BP were seen in the Normal Saline group at induction and after intubation. However, patients in Dexmedetomidine and Magnesium Sulphate groups had significantly lower HR, SBP and DBP at laryngoscopy and intubation. They concluded that BIS levels 40-50 ( $\pm 5$ ) there was no pressor response to intubation in the NS Group. Dexmedetomidine and Magnesium sulphate significantly reduced the heart rate and blood pressure from baseline. [37]

Pathak et al in their study aimed to compare Inj, Dexmedetomidine 1 $\mu$ g/kg and Magnesium sulfate 50 mg/kg in attenuation of cardiovascular response to laryngoscopy and intubation. Their observation was that the mean fall in HR, SBP, DBP and MAP values at 2 and 5 minutes of intubation were statistically highly significant ( $p=0.000$ ) in Dexmedetomidine group as compared to Magnesium sulphate group. [38]

Vivek Arora in his study compared magnesium sulphate and dexmedetomidine for attenuation of vasopressor stress

response during laryngoscopy and endotracheal intubation. Their observation was in group Dexmedetomidine and magnesium sulphate heart rate decreased up to 4 minutes and later on started increasing till 10 minutes and then decreased up to 30 minutes. In Normal saline group, mean heart rate increased till 7 minutes then started decreasing till 30 minutes. The mean systolic blood pressure (SBP) decreased pre-operatively till 5 minutes in all groups and then started increasing but not crossed 1st value. The mean SBP was higher in normal saline group ( $P < 0.05$ ). In Dexmedetomidine group and magnesium sulphate group, DBP decreased till 4 minutes and then started increasing till 30 minutes whereas in normal saline group, it increased till 2 minutes then decreased till 30 minutes. The difference was significant ( $P < 0.05$ ). Their conclusion was that both magnesium sulphate and dexmedetomidine both are efficacious in significantly reducing the heart rate and blood pressure from baseline. [39]

Godhki P et al in their study compared effectiveness of intravenous MgSO<sub>4</sub> 30mg/kg & Dexmedetomidine (DEX) 1 mcg/kg in attenuating vasopressor stress response during laryngoscopy and endotracheal intubation to study effect on heart rate, blood pressure & record complication if any. In their study their observation was Magnesium sulphate is as effective as Dexmedetomidine to attenuate vasopressor stress response to laryngoscopy and endotracheal intubation. [40]

Thus, the results of present study showed the both dexmedetomidine as well as magnesium sulphate could be used for control of Hemodynamic response except for heart rate for which magnesium sulphate failed to provide a reasonable attenuating effect. Both the drugs had minimal side effects with no significant difference between the groups. [41]

Owing to a better response for all the parameters, use of dexmedetomidine is recommended. However, considering that a

change in protocol of administration of magnesium sulphate could have provided an equivalent effect, further studies are recommended with a variable time gap between administration of magnesium sulphate and intubation to find whether this change could improve the efficiency of the drug further and may help in reducing the immediate increase in heart rate as observed in present study.

### Conclusion

At the end of the study we come to the conclusion that:

- Success of all major and specialized surgical procedures depends on balanced anaesthetic techniques, with use of induction agents, muscle relaxants and endotracheal intubation with minimal hemodynamic disturbance to the patients. Endotracheal intubation is an essential component of general anaesthesia as it serves many purposes including maintenance of the patency of upper airway, proper ventilation, reduction of the risk of aspiration and delivery of the inhalational anaesthetic agents from anaesthesia machine to the patients through breathing circuits.
- Laryngoscopy and intubation, being noxious stimuli, incite remarkable sympathetic activity. The pressor response, represented by an abrupt rise in the arterial blood pressure and heart rate (HR), arising 30 seconds after laryngoscopy and intubation, returns to baseline values steadily within 5–10 minutes.
- Incidence and level of hemodynamic changes are significant in susceptible patients, particularly those with systemic hypertension, coronary artery disease, valvular heart disease, cerebrovascular disease, and intracranial aneurysm, where even these transient changes can result in potentially deleterious effects e.g, acute left ventricular failure, arrhythmia,

myocardial ischemia, rupture of cerebral aneurysm. Traditionally used drugs like lignocaine, fentanyl, esmolol etc. are either not fully effective or may results in considerable adverse effects.

### Declarations:

**Funding:** None.

**Availability of data and material:** Department of Anaesthesiology & critical care. F.H. Medical College & Hospital, Etmadpur Agra.

**Code availability:** Not applicable.

**Consent to participate:** Consent taken.

**Ethical Consideration:** There are no ethical conflicts related to this study.

**Consent for publication:** Consent taken.

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