**Appendix 1. Detailed search strategies of PubMed, Scopus, EMBASE, and Web of Science was to capture relevant published studies of nebulized anticoagulation regimens as treatment for smoke inhalation associated acute lung injury.**

PUBMED :

(aerosols[mesh] OR nebulizers and vaporizers[mesh] OR administration, inhalation[mesh] OR nebulize[tiab] OR nebulized[tiab] OR nebulizer[tiab] OR nebulizers[tiab] OR nebulization[tiab] OR aerosol[tiab] OR aerosols[tiab] OR aerosolized[tiab] OR inhale[tiab] OR inhaled[tiab] OR inhalation[tiab] OR inhalable[tiab] OR vaporize[tiab] OR vaporizer[tiab] OR vaporizers[tiab]) AND (heparin[mesh] OR anticoagulants[mesh] OR antithrombins[mesh] OR heparin[tiab] OR anticoagulant[tiab] OR anticoagulants[tiab] OR anticoagulation[tiab] OR antithrombin[tiab] OR antithrombins[tiab]) AND (smoke inhalation injury[mesh] OR burns, inhalation[mesh] OR smoke[tiab] OR burn[tiab])

SCOPUS :

TITLE(nebulize OR nebulized OR nebulizer OR nebulizers OR nebulization OR aerosol OR aerosols OR aerosolized OR inhale OR inhaled OR inhalation OR inhalable OR vaporize OR vaporizer OR vaporizers) AND TITLE(heparin OR anticoagulant OR anticoagulants OR anticoagulation OR antithrombin OR antithrombins) AND TITLE(smoke))

SCOPUS :

(ABS(nebulize OR nebulized OR nebulizer OR nebulizers OR nebulization OR aerosol OR aerosols OR aerosolized OR inhale OR inhaled OR inhalation OR inhalable OR vaporize OR vaporizer OR vaporizers) AND ABS(heparin OR anticoagulant OR anticoagulants OR anticoagulation OR antithrombin OR antithrombins) AND ABS(smoke))

SCOPUS :

(TITLE(nebulize OR nebulized OR nebulizer OR nebulizers OR nebulization OR aerosol OR aerosols OR aerosolized OR inhale OR inhaled OR inhalation OR inhalable OR vaporize OR vaporizer OR vaporizers) AND ABS(heparin OR anticoagulant OR anticoagulants OR anticoagulation OR antithrombin OR antithrombins) AND ABS(smoke))

SCOPUS :

(ABS(nebulize OR nebulized OR nebulizer OR nebulizers OR nebulization OR aerosol OR aerosols OR aerosolized OR inhale OR inhaled OR inhalation OR inhalable OR vaporize OR vaporizer OR vaporizers) AND TITLE(heparin OR anticoagulant OR anticoagulants OR anticoagulation OR antithrombin OR antithrombins) AND ABS(smoke))

WEB OF SCIENCE :

Title=(nebulize OR nebulized OR nebulizer OR nebulizers OR nebulization OR aerosol OR aerosols OR aerosolized OR inhale OR inhaled OR inhalation OR inhalable OR vaporize OR vaporizer OR vaporizers) AND Title=(heparin OR anticoagulant OR anticoagulants OR anticoagulation OR antithrombin OR antithrombins) AND Title=(smoke)

WEB OF SCIENCE :

Author=(desai) AND Topic=(heparin OR anticoagulant OR anticoagulants OR anticoagulation OR antithrombin OR antithrombins) AND Topic=(nebulize OR nebulized OR nebulizer OR nebulizers OR nebulization OR aerosol OR aerosols OR aerosolized OR inhale OR inhaled OR inhalation OR inhalable OR vaporize OR vaporizer OR vaporizers)

EMBASE :

'aerosol'/exp OR 'nebulizer'/exp OR 'vaporizer'/exp OR 'inhalational drug administration'/exp AND ('heparin'/exp OR 'anticoagulant agent'/exp OR 'antithrombin'/exp OR 'anticoagulant therapy'/exp) AND 'lung burn'/exp

EMBASE :

nebulize:ti OR nebulize:ab OR nebulized:ti OR nebulized:ab OR nebulizer:ti OR nebulizer:ab OR nebulizers:ti OR nebulizers:ab OR nebulization:ti OR nebulization:ab OR aerosol:ti OR aerosol:ab OR aerosols:ti OR aerosols:ab OR aerosolized:ti OR aerosolized:ab OR inhale:ti OR inhale:ab OR inhaled:ti OR inhaled:ab OR inhalation:ti OR inhalation:ab OR inhalable:ti OR inhalable:ab OR vaporize:ti OR vaporize:ab OR vaporizer:ti OR vaporizer:ab OR vaporizers:ti OR vaporizers:ab AND (heparin:ti OR heparin:ab OR anticoagulant:ti OR anticoagulant:ab OR anticoagulants:ti OR anticoagulants:ab OR anticoagulation:ti OR anticoagulation:ab OR antithrombin:ti OR antithrombin:ab OR antithrombins:ti OR antithrombins:ab) AND ('smoke inhalation' OR 'lung burn'/exp)

**Appendix 2 . Inhaled anticoagulation regimens in ovine models of smoke inhalation associated acute lung injury.**

| Author (Year); Reference # | Design (N) | Intervention | Primary Endpoints | Other Endpoints / Comments |
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| Desai et al. (1986);(32)* | Prospective, randomized, controlled (33) | 1. Control untreated (n=7)<br>2. 90% DMSO 3ml INH (n=7)<br>3. 90% DMSO with Heparin 10,000 units INH (n=7)<br>4. Heparin 10,000 units INH (n=6)<br>5. NAC 5ml INH (n=5).<br><br>Drugs were nebulized every 4 hours for 48 hours. | Survival at 48 hours:<br>1. Controls: 0% (0/7)<br>2. DMSO: 43% (3/7)<br>3. DMSO + Heparin: 100% (7/7) with 7 weaned from ventilator<br>4. Heparin: 67% (4/6) with 1 weaned from ventilator<br>5. NAC: 100% (6/6) with 6 weaned from ventilator | ACT not altered by heparin administration. |
| Brown et al. (1988);(33) | Prospective, Randomized, Placebo Controlled (26) | 1. Control untreated (=7)<br>2. 90% DMSO 3ml (n=6)<br>3. Heparin 10,000 units (n=7)<br>4. Heparin 10,000 units + 90% DMSO 3ml (n=6).<br><br>Nebulized treatments administered every 4 hours for 48 hours post-injury. Sheep were studied for 96 hours. | Survival at 96 hours:<br>1. Control 0% (0/7)<br>2. DMSO 33% (2/6)<br>3. Heparin 66% (4/6)<br>4. Heparin and DMSO 100% (7/7)<br>$P < 0.05$ compared to controls and DMSO alone. | Due to high mortality in control and DMSO groups, further comparisons were only made between Heparin and DMSO + Heparin groups. ↓ Lung lymph flow in the DMSO + Heparin group. ↓ respiratory rates in heparin group to maintain PaCO2 , but ↑ peak airway pressures compared to DMSO + Heparin group. ACT not altered by heparin administration. |
| Cindrick et al. (1996);(34)* | Prospective, randomized, placebo controlled (12) | 1. Control: Lactated Ringer's 3ml INH (n=6)<br>2. Heparinoid analog GM-1892 2mg/kg INH (n=6)<br><br>Solutions were delivered via in-line nebulization 1 hour post-injury and then every 4 hrs for 48hrs. | GM-1892 attenuated the increase in lymph flow when compared to controls ($P < 0.05$) | |
| Katahira et al. (2001);(35)* | Prospective, randomized, placebo controlled (16) | 1. Heparin 10,000 units INH (n=5)<br>2. Saline 0.9% INH (n=5)<br>3. Sham, non-treatment group (n=6)<br><br>Nebulized treatments were administered 1h post-injury and then every 4 hours for 24 hours. | PaO2/FiO2 at 24 hours:<br>1. Heparin: 287.8 ± 29.1<br>2. Saline: 192.2 ± 39.4 | The post-injury ↑ lung lymph was significantly attenuated in the heparin group. Plasma AT-III levels ↓ significantly in all groups ($p<0.05$). |
| Suman et al. (2001);(36)* | Prospective, randomized, placebo controlled (10) | 1. Heparin 10,000 units INH<br>2. Saline 5ml<br><br>Nebulized treatments administered every 4 hours post-injury. | $C_{dyn}$ was significantly ↑ at all time points in the heparin group.<br>$R_{AW}$ was significantly ↓ in the heparin group at 30 hours, but unchanged at 24 & 48 hours.<br>WOB was significantly ↓ at all time points in the heparin group. | PaO2/FiO2 did not differ significantly between groups at any time point. |

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| Murakami et al. (2002);(65) | Prospective, randomized, with sham and placebo controls (19) | 1. Heparin 10,000 IU INH (n=5)<br>2. Heparin 300 IU/kg/23h IV (n=5)<br>3. Saline INH (n=5)<br>4. Sham, non-injured (n=4)<br><br>Animals also inoculated with pseudomonas $5x10^{11}$ CFU. All treatments initiated 1 hour post-injury. Nebulized treatments administered every 4 hours. | Gas exchange at 24 hours was improved in heparin INH group as marked by attenuated changes in PaO2/FiO2, $Q_S/Q_T$, and W/D. | ↓ Histological obstruction in heparin INH group. Non-significant trend toward ↑ pulmonary arterial and left atrial pressures, and cardiac index. No change in plasma nitric oxide levels. Attenuated fall is systemic vascular resistance in heparin INH group. |
| Taski et al. (2002);(49) | Prospective, randomized, placebo control (18) | 1. Saline INH (n=6)<br>2. Heparin 10,000 IU INH (n=6)<br>3. Heparin 10,000 IU INH + lisophylline 20mg/kg IV bolus then 10 mg/kg/h IV continuous infusion (n=6)<br><br>Treatments were initiated 30 minutes post-exposure and continued for 48 hours. Nebulized treatments administered every 4 hours. | Degree of respiratory failure at 48 hours was determined by assessing requirements for (a) mechanical ventilation, (b) supplemental O2, and (c) a-A gradient.<br>1. Saline: (a) 4/6; (b) 5/6<br>2. Heparin: (a) 1/6; (b) 2/6)<br>3. Heparin + Lisophylline IV: (a) 0/6 (p<0..05); (b) 0/6<br>Group 3 showed attenuation in rise of a-A gradient. | The ↓ Qs/Qt seen in groups 2 & 3 was only statistically significant for group 3. No significant change in W/D was observed. No significant change in myeloperoxidase activity or malondialdehyde was observed in the group 2, however significantly ↓ values observed in Group 3 (p<0.05). No change in histology scores was reported. |
| Thomas et al. (2004);(38)* | Prospective, randomized, placebo controlled (12) | 1. Sham, non-injured, non-treated (n= 3)<br>2. Saline INH (n=3)<br>3. TPA 1mg INH (n=3)<br>4. TPA 2mg INH (n=3) treated groups.<br><br>Nebulized treatments initiated at 4 hours post-injury and continued every 4 hours for 48 hours. | DLCO (ml/min/mmHg) was measure at (a) baseline and (b) 48 hours:<br>1. Sham: (a) 26 ± 6; (b) 24 ± 4<br>2. Saline: (a) 25 ± 4; (b) 9.6 ±1.6<br>3. TPA 1mg: (a) 18 ± 4; (b) 8.3 ± 1.6<br>4. TPA 2mg: (a) 20 ± 4; (b) 20 ± 2 | |
| Enkhbaatar et al. (2004);(17)* | Prospective, randomized, placebo and sham controlled (26) | 1. Sham non-injured, non-treated (n=6)<br>2. Saline INH (n=5)<br>3. Heparin 10,000 IU INH (n=5)<br>4. AT 290 units INH (n=5)<br>5. Heparin 10,000 IU INH + AT 290 units INH (n=5)<br><br>Nebulized treatments administered every 4 hours for 48 hours. | Sham animals remained stable. Controls showed a marked ↓ PaO2/FiO2, ↑ pulmonary shunt, ↑ lung lymph flow, ↑ pulmonary vascular permeability, and ↑ lung wet-to-dry ratio. These findings were associated with increased airway obstruction and airway pressures. These findings were attenuated by combined Heparin + AT but not heparin alone or AT | |
| Enkhbaatar et al. (2005);(39)* | Prospective, randomized, placebo controlled | 1. Sham, non-injured, non-treated<br>2. Saline INH<br>3. TPA 2mg INH<br>4. AT 290 units INH + heparin 10,000 units INH<br><br>Nebulized treatments initiated 2 hours post-injury and continued every 4 hours for 48 hours. | At 48 hours measurements were made of (a) bronchial obstruction score (%), (b) lung water content, (c) lung lymph flow (ml/h).<br>1. Saline: (a) 19±3; (b) 5.7±0.2; (c) 50±3<br>2. TPA: (a) 12±2†; (b) 4.7±0.3†; (c) 12±4† | |

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| | | | 3. AT + Heparin: (a) 11±2.5†; (b) 4.7±0.2†; (c) 29±11<br>†p < 0.05 vs. saline. | |
| Nakano et al. (2006);(40) * | Prospective, randomized, placebo control (14) | 1. Sham, non-injured, non-treated (n=6)<br>2. Saline 10ml INH (n=4)<br>3. Heparin 10,000 IU, 10ml, INH + rhAT 0.34mg/kg/h IV (n=4)<br><br>Live P. *aeruginosa* ($1.5\text{-}3x10^{11}$ CFU) instilled into airways. All treatments initiated 2 hours post-injury and continued for 24 hours. Nebulized treatments administered every 4 hours. | Markers of pulmonary gas exchange and congestion were assessed at 24 hours including: (a) PaO2/FiO2 and (b) Qs/Qt.<br>1. Sham: (a) 619.2± 6.7; (b) 0.13±0.1<br>2. Saline: (a) 81.3±21.8*; (b) 0.61±0.1*<br>3. Heparin: (a) 177.4±24.3†; (b) 0.37±0.05†<br>*p < 0.05 vs. sham,; † p < 0.05 vs. saline control. | |
| Enkhbaatar et al. (2007);(41) | Prospective, randomized, with placebo and sham controls (25) | 1. Sham, non-injured and non-treated (n=6)<br>2. Saline 15 ml control INH (n=6)<br>3. rhAT 290 units (5ml) INH + heparin 10,000 IU (10ml) INH (n=6)<br>4. rhAT 290 units (5ml) INH (n=5)<br>5. Heparin 10,000 IU (10ml) INH (n=5)<br><br>Treatments initiated 2 hours post-injury and continued every 4 hours for 24 hours. | Airway obstruction scores for groups 4 and 5 significantly improved compared to group 2, but did not differ significantly between each other. The rhAT + Heparin group displayed improved PIP and $P_{PL}$ compared to groups 1 & 2. Groups 3, 4, 5 demonstrated improved $PaO_2/FiO_2$. Group 3 displayed improved Qs/Qt and lung lymph flow compared to groups 1 and 2.<br><br>* Note: not all variables reported for each group | Treatments attenuated fall in serum AT, but did not affect ACT. |
| Enkhbaatar et al. (2008);(42) | Prospective, randomized, placebo & sham controlled (18) | 1. Sham, non-injured, non-treated (n=6)<br>2. Saline group (n=6(42))<br>3. rhAT 0.34 mg/h IV continuous infusion + heparin 10,000 IU INH<br><br>Treatments initiated 1 hour post-injury and continued for 48 hours. Nebulized treatments administered every 4 hours. | Treatment with rhAT + heparin attenuated rise in PIP and $P_{PL}$ as well as attenuated ↑ lung lymph flow and W/D. | rhAT + heparin attenuated fall in serum AT levels. rhAT + heparin attenuated ↑ in MPO, NO, and VEGF. |
| Rehberg et al. (2009);(43) | Prospective, randomized, placebo controlled (8) | 1. rhAT III IV infusion at 6 U/kg/h + heparin 10,000 IU INH + TPA 2 mg INH (n=4)<br>2. Saline (n = 4)<br><br>rhAT, heparin, and TPA started at 1, 2, and 4 hours post-injury respectively. Nebulized treatments administered every 4 hours. | rhAT + heparin therapy was associated with:<br>↓ PaO2/FiO2 (p < 0.01)<br>↓ PIP (27 ± 1 cmH2O vs 37 ± 1 cmH2O; p < 0.001)<br>↓Qs/Qt (22 ± 2% vs 42 ± 4%; p = 0.004)<br>↓pulmonary lymph flow (38 ± 5 mL/h vs 54 ± 4 mL/h; p<0.05)<br>↓permeability index (18 ± 3 mL/h vs 26 ± 1 mL/h; p < 0.05). | No change in systemic coagulation parameters. rhAT + heparin attenuated fall in plasma AT (Treatment: 107 ± 5% vs Control: 69 ± 5% of baseline level; p = 0.001). |
| Asmussen et al. (2010);(44)* | Prospective, randomized, placebo | 1. rhAT 0.7mg/kg/h continuous IV infusion + heparin 5,000 IU INH + TPA 2mg INH (n=4)<br>2. Control (vehicle only) (n=4) | Survival at 96 hours was:<br>1. rhAT + heparin + TPA: 4/4<br>2. Control: 3/4 | The PaO2/FiO2 ratio was significantly improved in treatment group at 96h (391±96 vs. 214±16). (a) |

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| | controlled (8) | rhAT, heparin, and TPA started at 1, 4, and 4 hours post-injury respectively. Nebulized treatments administered every 4 hours for 48 hours. | | Ventilator weaning and (b) tracheostomy decannulation at 96 hours was:<br>1. Treatment: (a) 4/4; (b) 3/4<br>2. Control: (a) 0/4; (b) 0/4 |
| Rehberg et al. (2010);(66)* | Prospective, randomized, placebo controlled (16) | 1. Control: Saline IV (n=6)<br>2. rhAT III 6 u/kg/h IV infusion (n=6)<br>3. rhAT III 6 u/kg/h IV infusion + heparin 10,000 IU INH + TPA 2mg INH (n=4)<br>rhAT III, heparin, and TPA started 1, 2, and 4 hours post-injury. Nebulized treatments administered every 4 hours for 48 hours. | At 48 hours, both strategies improved PaO2/FiO2 (control: 83 ± 6 mmHg; rhATIII: 199 ± 48 mmHg; combination: 360 ± 29 mmHg; $p < 0.05$ each). Combination therapy was associated with significantly ↓ pulmonary vascular resistance (119 ± 6 vs. 166 ± 17 dyn s cm-5 m-2 ) and Qs/Qt (22 ± 2 vs. 30 ± 4%) as compared to rhAT III alone. rhAT III mono-therapy resulted in a greater ↓ pulmonary lymph flow (16 ± 7 vs. 38 ± 5 mL h-1). | Neutrophil migration ↓ only in rhAT III infusion (control: 330 ± 64% of baseline; rhAT III: 50 ± 22% of baseline; combination: 330 ± 94% of baseline). rhAT III single-therapy ↓ net fluid balance. |
| Rehberg et al. (2010 & 2011);(47, 67)* | Prospective, randomized, placebo controlled (18) | 1. Control: Saline INH (n=6)<br>2. rhAT III 6 u/kg/h IV infusion + TPA 2mg INH + heparin 5,000 IU INH (n=6)<br>3. rhAT III 6 u/kg/h IV infusion + TPA 2mg INH + heparin 10,000 IU INH (n=6)<br><br>rhAT III, TPA, and heparin started 1, 4, and 4 hours post-injury. Nebulized treatments administered every 4 hours. Therapy continued for 48 hours. | Lung injury was assessed at 48 hours via: (a) PaO2/FiO2; (b) PIP ($cmH_2O$)<br>1. Control: (a) 134±30; (b) 36±2<br>2. Low dose heparin: (a) 276±44; (b) 27±2<br>3. High dose heparin: (a) 352±25; (b) 27±1 | Low dose heparin group exhibited lower transvascular fluid flux, permeability index, net fluid balance and higher serum protein levels. |
| Asmussen et al. (2011);(48)* | Prospective, randomized, placebo controlled (10) | 1. rhAT 0.7 mg/kg/hour IV infusion + heparin 5,000 IU INH + TPA 2 mg INH<br>2. Saline INH<br><br>rhAT, heparin, and TPA started at 1, 2, and 4 hours post-injury respectively. Nebulized treatments administered every 4 hours for 48 hours. | Survival at 96 hours:<br>1. rhAT + heparin + TPA: 5/5<br>2. Control: 3/5 | Respiratory insufficiency at 96 hours as measured by (a) PaO2/FiO2; (b) wean from MV; (c) tracheostomy decannulation was:<br>1. Control: (a) 267±51; (b) 0/5; (c) 0/5<br>2. Treatment: (a) 530±16; (b) 5/5; (c) 4/5 |

* Published as abstract.

IV - Intravenous; INH - inhaled; ACT - activated clotting time; DMSO - dimethylsulfoxide; $C_{dyn}$ - dynamic compliance; $R_{AW}$ – airway resistance; WOB - work of breathing; aA-gradient - alveolar-to-arterial gradient; W/D - Wet-to-dry ratio; Qs/Qt – shunt fraction; AT - antithrombin; rhAT - recombinant human antithrombin; PIP - peak inspiratory pressure; $P_{PL}$ – plateau pressure; TPA - tissue plasminogen activator; DLCO - diffusion capacity of carbon monoxide; NO - nitric oxide