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PRE-CLINICAL STUDIES OF INTERMITTENT MODULAR PLASMA ADSORPTION OF CYTOKINES AND TOXINS (IMPACT SYSTEM) FOR ARTIFICIAL LIVER SUPPORT

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Abstract

Purpose of Study: Episodic type C hepatic encephalopathy (HE) developing in decompensated cirrhosis carries considerable morbidity and mortality. Our aim was to investigate the in vitro clearances and in vivo safety of an artificial liver support system developed to treat liver failure and HE (IMPACT System™ with HLM-100 Adsorption Column and HLM-200 Plasma Separator).

Methods: The IMPACT System™ consists of continuous plasma filtration utilizing a plasma filter (HLM-200 Plasma Separator) and sorbent adsorbent detoxification of plasma utilizing both charcoal and nonionic resins within an adsorption column (HLM-100 Adsorption Column). Each of the resins within the column was studied individually, in addition to various combinations of the three resins. Initially we investigated the system's clearances using 3 liters of expired heparinized human plasma with target concentrations of bilirubin (20 mg/dl), urea nitrogen (50 mg/dl), creatinine (5 mg/dl), and acetaminophen (175 mcg/ml). This plasma was circulated through the IMPACT System™ for 4 hours (column processed 6 liters of plasma filtrate). The final combination of resins in the IMPACT System™ removed significant amounts of total bilirubin ($34.6 \pm 2.1\%$, $p < 0.05$), urea nitrogen (26.3 ± 1.2 , $p < 0.01$), creatinine (76.8 ± 0.1 , $p < 0.001$) and acetaminophen ($80.5 \pm 1.0\%$, $p < 0.001$). There were only modest decreases in serum proteins ($9.5 \pm 0.4\%$, $p < 0.05$), albumin ($7.8 \pm 0.2\%$, $p < 0.05$), and fibrinogen ($22.5 \pm 1.5\%$, $p < 0.01$) without effect on serum electrolytes. Secondly, eight approximately 55 pound anesthetized and mechanically ventilated dogs were treated with the IMPACT System™ with heparin anticoagulation. Following an 1-hour lead-in phase with the circuit excluding the HLM-100 Adsorption Column, 4-hour treatments with the adsorption column included flow rates intended for adult humans. Monitoring included laboratory testing and pulmonary artery pressure and continuous arterial measurements. Testing demonstrated that inclusion of the HLM-100 Adsorption Column and HLM-200 Plasma Separator into an extracorporeal circuit did not result in evidence of hemodynamic instability (mean arterial pressure 70 ± 7 to 92 ± 5 mmHg, $p < 0.05$), hemolysis (hematocrit 42 ± 4 to 36 ± 2 , $p = \text{NS}$), thrombocytopenia (platelets 132 ± 11 to

104±8, p=NS), leucopenia (leukocytes 5.6±0.8 to 7.9±1.3, p<0.05), electrolyte derangements, or nonspecific loss of fibrinogen (52±8 to 57±5 mg/dL, p=NS) or albumin (1.4±0.2 to 1.3±0.1 g/dL, p=NS).

Conclusion: The IMPACT System™ achieves excellent in vitro clearances of common liver failure, kidney failure and exogenous toxins and was safe and well tolerated in healthy dogs without signs of bio-incompatibility or adverse effects on hemodynamic parameters, electrolytes, or serum proteins.

Introduction

An estimated 5.5 million Americans have cirrhosis (prevalence ~2000 per 100,000 population) with ~20% per year developing hepatic encephalopathy (HE) resulting in ~250,000 hospital admissions. HE associated with hepatic failure is attributed to hepatic insufficiency with impaired detoxification and passage of toxic nitrogenous substances from portal to systemic circulation. Manifestations range from slowed mentation and confusion to respiratory failure and coma. The presence of HE nearly triples the relative risk of death (RR 2.71) among patients with cirrhosis and ascites. Only half of patients will survive an additional 1-2 years. Artificial liver support with devices such as sorbent-based detoxification have a positive effect on HE and decreases mortality ~33% (OR 0.67, 95% CI 0.51-0.90) in acute-on-chronic liver failure (JAMA 2003;289:217-22). Biocompatibility and anticoagulation issues have limited the clinical effectiveness of currently available devices. The development of the IMPACT System™ with a plasma separator (0.45 micron) that creates an environment for detoxification similar to that found in the liver holds tremendous promise as a safe and effective artificial liver support device [**HemoLife Medical Inc. 1231 Puerta del Sol, Suite 500, San Clemente, CA. Phone: (949) 498-9461 www.hemolifemedical.com**].

Methods

Initial in vitro studies: We investigated the system's clearances using 3 liters of expired heparinized human plasma [target concentrations of bilirubin (20 mg/dl), urea nitrogen (50 mg/dl), creatinine (5 mg/dl), and acetaminophen (175 mcg/ml)] circulated through the IMPACT System™ for 4 hours (column processed 6 liters of plasma filtrate).

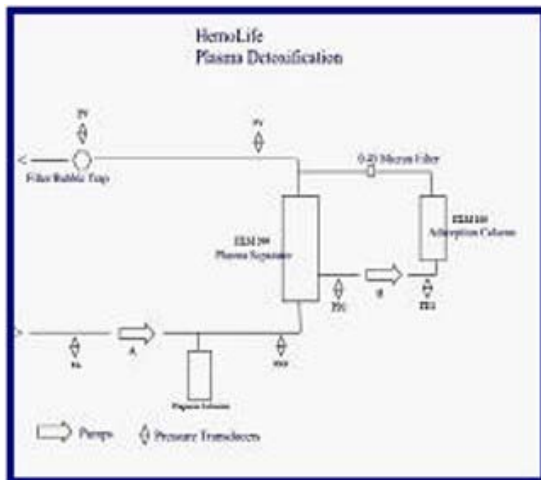
Subsequent canine studies: Eight (8) healthy approximately 55 pound mongrel dogs were sedated, intubated, and placed under general anesthesia. A 11.5 Fr standard dialysis catheter (Ash-Split, MedComp) was placed in right internal jugular vein. Heparin anticoagulation was used with measurements of whole blood activated coagulation time (ACT Hemachron, International Technidyne Corporation) used to monitor therapy with a goal of maintaining the ACT of 300 seconds.

Following an 1-hour lead-in phase excluding the HLM-100 Adsorption Column (plasma flow rate 0 ml/min), 4-hour treatments including the complete IMPACT System™ (HLM-200 Plasma Separator and HLM-100 Adsorption Column) were performed. The IMPACT System™ utilizes the B|Braun DIAPACT® CRRT machine in the plasma adsorption and perfusion (PAP) mode. The dogs' blood was circulated through the HLM-200 Plasma Separator (polysulfone, 0.45 micron, 1.0 m²) to generate the plasma filtrate that passes through the HLM-100 Adsorption Column (sorbents included a combination of charcoal and biocompatible nonionic resins). The flow rates on the blood (Figure 1. A-125 mL/min) and plasma (Figure 1. B-25 mL/min) limbs

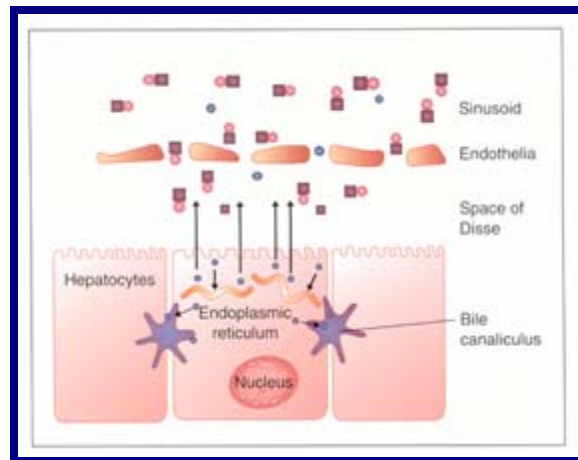
were those intended for adult human treatments. The 4-hour treatments with the IMPACT System™ allows 6 L of plasma filtrate to pass through the HLM-100 Adsorption Column.

The IMPACT System™ circuit pressures were monitored utilizing the internal software of the BJBraun DIAPACT® CRRT machine.

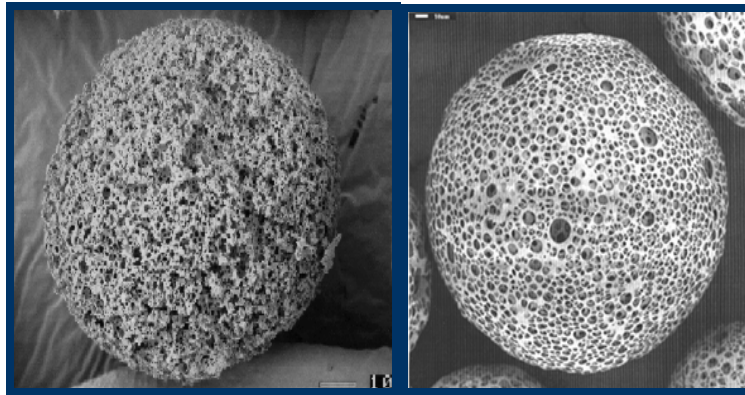
Hemodynamic parameters were followed with a pulmonary arterial (central venous and pulmonary arterial systolic, diastolic and mean pressures, cardiac output and systemic vascular resistance) and peripheral arterial catheters (systolic, diastolic, and mean arterial pressures). Urine output was monitored with an urinary catheter. The complete blood cell counts, arterial blood gases, and concentrations of sodium, potassium, chloride, CO₂, urea nitrogen, creatinine, glucose, total protein, albumin, calcium, total/direct bilirubin, AST/ALT, and acetaminophen were at T-60, T-30, 0, 15, 30, 60, 120, 180, and 240 minutes.



Normal Liver Demonstrating Plasma Separation Effect of Sinusoidal Fenestrations



Nonionic Resins in Adsorption Column Demonstrating Porous Nature



Results

In vitro Studies:

Figures 1 & 2 summarize in vitro clearance rates of HLM-100 Adsorption Column for common endogenous toxins (bilirubin $34.6 \pm 2.1\%$ $p < 0.05$, urea nitrogen 26.3 ± 1.2 $p < 0.01$, and creatinine 76.8 ± 0.1 $p < 0.001$), and an exogenous toxin (acetaminophen $80.5 \pm 1.0\%$, $p < 0.00$).

Healthy Canine Studies:

Treatments were well tolerated with no hemodynamic instability (mean arterial pressure pre 70 ± 7 vs post 92 ± 5 mmHg, $p < 0.05$) or significant change in serum electrolytes (data not shown).

The B|Braun DIAPACT CRRT monitoring demonstrated excellent stability in measured pressures within the IMPACT System™ circuit.

Table 1 summarizes the overall biocompatibility of the IMPACT System™ with respect to effect on blood levels of platelets, hemoglobin, leukocytes, and serum levels of total protein, albumin, and fibrinogen. All results given \pm SD.

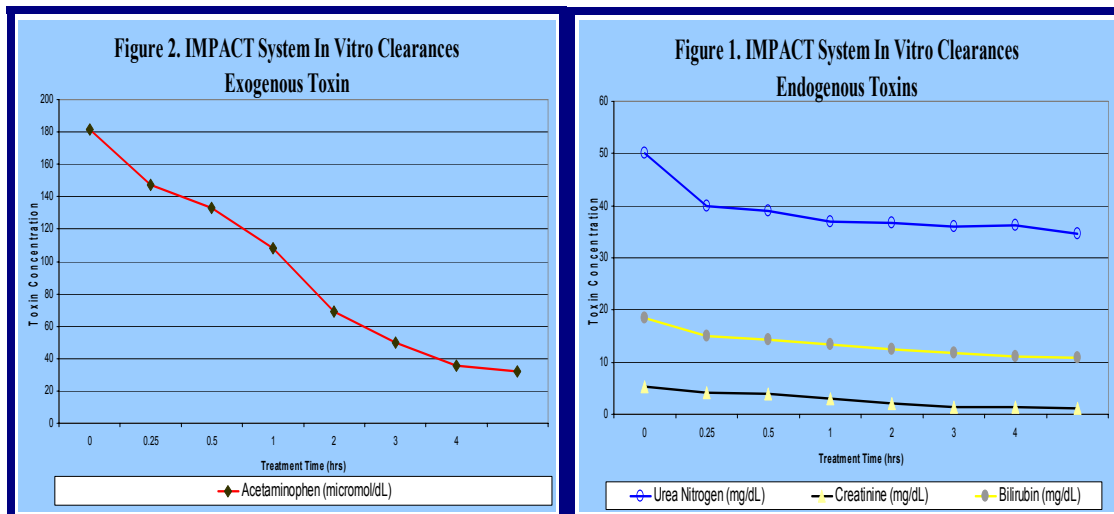


TABLE 1. IMPACT System™ Biocompatibility Measures

<u>Value</u>	<u>T=0 mins</u>	<u>T=240 mins</u>	<u>p</u>
Hemoglobin (g/dL)	14.8±4.0	12.6±2.0	NS
Platelet (Thousand/mL)	104±23	93±24	NS
WBC (Thousand/mL)	4.0±1.5	7.9±3.8	<0.05
Serum Fibrinogen (mg/dL)	52±22	57±5	NS
Serum Albumin (g/dL)	1.4±0.4	3±0.4	NS

Conclusions

The IMPACT System™ achieves excellent in vitro clearances of common liver failure, kidney failure and exogenous toxins and was safe and well tolerated in healthy dogs without signs of bio-incompatibility or adverse effects on hemodynamic parameters, electrolytes, or serum proteins.

An FDA-approved IDE pilot study investigating the IMPACT System™ in patients with decompensated cirrhosis and hepatic encephalopathy is in progress.

For More Information Contact:

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