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Haemoglobin solutions – where are we going?

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Haemoglobin solutions intended to transport oxygen in the plasma were first conceived and developed as an alternative to red cell transfusion. Although many different haemoglobin formulations have been manufactured and submitted to extensive clinical trials in humans, most of these products have now been abandoned because of safety concerns; only the Biopure product, Hemopure, remains available for clinical use.

HBOC-201 (Hemopure) is a cell-free, polymerised haemoglobin solution that, when infused intravenously, has the following physiological and pharmacological actions:

- The haemoglobin polymer carries oxygen in the plasma in a similar manner to normal red cell haemoglobin, i.e. each gram of haemoglobin carries 1.34 ml of oxygen.[1-3]
- In addition, the presence of haemoglobin molecules in the plasma enhances the efficiency of both the uptake of oxygen at the pulmonary alveolar level, and the off-loading of oxygen in the tissue capillaries by the host red blood cells.[4]
- This enhanced efficiency of oxygen exchange improves the delivery of oxygen to the tissues, resulting in an increased tissue oxygen tension.[5,6]

HBOC-201 is manufactured from a plentiful and well-controlled source, using cattle from an extensive herd-management programme as donors to ensure that only certified disease-free animals provide the bovine haemoglobin. Extraction and purification processes produce a sterile, pyrogen-free solution containing glutaraldehyde cross-linked bovine haemoglobin polymers, with an average molecular weight of 250 kD, in a balanced salt solution.

This haemoglobin polymer has an oxygen dissociation curve that is right-shifted with a P_{50} of 43 mmHg, compared with 27 mmHg for human haemoglobin.[2,3] The oxygen affinity of human haemoglobin relies on adequate levels of 2,3-diphosphoglycerate compared with that of bovine haemoglobin, which is regulated by the concentration of chloride ions in the plasma. It has a dose-dependent intravascular half-life of 16 - 24 hours. When stored between 2°C and 30°C, it is stable for at least 3 years, and can be infused directly without any prior reconstitution. Laboratory typing and cross-matching are not required because haemoglobin solutions are cell free, and there are no cell surface antigens present.

Bovine-derived haemoglobin solution (HBOC-201) was first registered for routine clinical use in South Africa for the treatment of adult surgical anaemia. This product has been administered to nearly 500 patients since registration, initially as part of a clinical surveillance and educational programme and subsequently in commercial sales, with ongoing surveillance of safety information.

Avoidance of blood was achieved in over 90% of patients when HBOC-201 was used as an alternative to conventional red cell transfusion.[7] HBOC-201 adequately oxygenated and stabilised patients as demonstrated by clinical behaviour, and the vital parameters were monitored during the infusion. In no instance was it required to supplement HBOC-201 infusions with red blood cell transfusion for reasons of inadequate, initial clinical response. As clinicians have become more expert in the clinical use of HBOC-201 in South Africa, so the adverse effect profile has been more favourable, with a product-related serious side-effect rate of approximately 2 per 100 most recent patient exposures.

Management of acute anaemia when red cells are not an immediate option

The administration of HBOC-201 has in many cases proved to be life saving, particularly when human red cells are not an immediate option in the treatment of severe and acute anaemia.[7] Currently, the cost of developing and manufacturing any haemoglobin solution, and the relatively short half-life of the currently available products, restrict these drugs from being considered an everyday alternative to conventional red cell transfusion. However, when patients experience sudden, unexpected blood loss and when red cells are not immediately available, a haemoglobin solution that has a long, stable shelf-life, does not require refrigeration, and does not need cross-matching can be life saving. Other scenarios where red cells are not an option include religious beliefs, in allo-immunised patients, and in patients suffering from acute auto-immune haemolytic anaemia. HBOC-201 has been used with great success in these categories of patients.[7] Currently, other than a red cell transfusion, no available therapeutic measure offers the acutely anaemic patient an immediate and effective method of oxygen transport and delivery.

Increased tissue oxygen levels have been demonstrated after the administration of a haemoglobin-based oxygen carrier.[5,6] This finding raises the possibility that this class of pharmacological agent can be used in the future treatment of acutely ischaemic tissues such as seen in acute myocardial, limb and gut ischaemia. In animal models of acute limb ischaemia[6] and acute coronary ischaemia,[8] improved tissue oxygenation at the site of reduced blood flow and tissue-protective effects have been unequivocally demonstrated. Certainly, anecdotal cases where HBOC-201 has been used effectively as a tissue oxygenator for relief of acute ischaemic symptoms in acute coronary, limb and gut ischaemia support this concept. The role of haemoglobin solutions as enhancers of tissue oxygen delivery and their effect on acute tissue ischaemia and wound healing are currently the subject of prospective randomised controlled studies.

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Cell-salvage techniques – a practical approach

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Blood is an expensive and increasingly scarce commodity. Moreover, the transfusion of homologous blood carries the risk of major transfusion reactions and transmission of infective agents such as the hepatitis and HI viruses. Therefore, there is a worldwide resistance to homologous blood transfusion, and a policy of bloodless surgery is becoming increasingly popular.

For the past 13 years efforts have been made to limit the use of blood transfusion in the open-heart unit at Greenacres Hospital, Port Elizabeth. This article gives an overview of the techniques used to salvage and transfuse red blood cells during cardiac surgery. This policy results in the haematocrit being at an optimal level during cardiopulmonary bypass surgery, allowing the majority of patients to return to the ICU without having received a transfusion of homologous blood. Cell saving has emerged as an indispensable part of our strategy. An added bonus of cell saving is that inflammatory mediators are removed during the cell-saving process, reducing the systemic inflammatory response associated with conventional cardiopulmonary bypass surgery.

Briefly, the techniques we follow are:

1. Minimising blood loss during surgery.
2. Intraoperative cell salvage with the Electra 5 cell saver.
3. Postoperative cell salvage in the ICU when indicated (blood loss more than 500 ml in the first 2 hours postoperatively).

Minimising blood loss during surgery

This is the most important factor in conserving autologous blood. In our experience excessive blood loss during the procedure often goes unnoticed, collecting in open pleural spaces.

Sources include bleeding from unsecured side branches of the internal mammary artery, the internal mammary bed, the internal periosteum of the sternum, the arteries around the xiphisternum, the leg after vein harvesting and cannulation sites. We take particular care to ascertain that all these areas are dry, and make liberal use of cautery and the argon laser technique.

Intraoperative cell salvage

A custom-designed sucker facilitates heparin being delivered to the operation site so that shed blood (which is immediately sucked away) is mixed with anticoagulant. Haemolysis is minimised by setting the vacuum at the lowest effective level. The blood/anticoagulant mixture carried into the sterile reservoir is filtered to remove large clots and debris. The blood must be anticoagulated to prevent clotting and secondary fibrinolysis.

Great care is taken to administer the correct amount of anticoagulant. A ratio of 1:5, and not more than 1:10, of anticoagulant to collected blood is recommended. Most collection reservoirs have filters in the 40 - 150 µm range.

Blood and anticoagulant are then drawn from the collection container and centrifuged. The red cells are separated from waste products and suspended in a sterile, isotonic saline solution. Waste products include white cells, platelets, plasma, anticoagulant, fats and free plasma haemoglobin. These products are collected in a bag and discarded. Packed red cells that have been separated from the waste products are collected separately. Then the washed red cells are reinfused into the circulation using a 40 µm leucocyte filter.

Postoperative cell salvage

When there is excessive bleeding into the underwater drainage bottles immediately after the operation the cell-saving machine is transferred, together with the patient, to the ICU, allowing blood from the bottles to be processed. Normal saline (not plain water) is placed in the underwater drainage bottles. Blood can be collected and washed up to 6 hours postoperatively.

Before 2001 only conventional cardio-pulmonary bypass procedures were done in our unit. The cell saver was used to recover red cells not recovered by the cardiotomy sucker. Cells were recovered from e.g. blood-soaked surgical swabs and very low haematocrit blood (in pleural spaces) judged unsuitable for direct return to the heart-lung machine because of haemodilution. However, most shed blood was still returned to the heart-lung machine via the cardiotomy sucker. Blood from the cardiotomy sucker contains elevated inflammatory markers and is an independent source of inflammatory mediators.

In 2001, with this in mind, we changed from the conventional cardiopulmonary bypass procedure to the mini bypass closed circuit. This method allows a low priming volume. Tip-to-tip coated tubing and the use of a centrifugal pump minimise blood damage. Using this method all the shed blood, including that removed by the cardiotomy sucker, is returned to the cell saver. Washed and suspended red cells are then returned to the open heart circuit.

Using the mini circuit, combined with the cell saver, the following observations were made:

- The haematocrit remained constantly high during cardiopulmonary bypass. During the procedure it was noted that the haematocrit dropped 4 - 5%. However, at the end of the procedure, with washed red blood cells having been reinfused, the haematocrit had usually returned to the preoperative value.
- The need for homologous blood transfusion diminished considerably. Currently, only 7% of patients require blood transfusion postoperatively.

Further reading:

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