Whole Blood Cardioplegia: Do We Still Need to Dilute?

Jakob Vinten-Johansen, PhD

Division of Cardiothoracic Surgery, Department of Surgery, Emory University, Atlanta, Georgia

Presented at the Advanced Myocardial Protection Symposium, San Antonio, Texas, October 9, 2015.

I have been tasked to talk about the topic “Whole Blood Cardioplegia: Do We Still Need to Dilute?” This is a question that was raised almost 20 years ago by Dr. Philippe Menasché (1) from Paris, and it’s interesting that we are still asking this question in 2015. We will get into the data that drive this persistent question during this discussion. This presentation will cover 1) a bit of history of blood cardioplegia, 2) the attributes of both hemodiluted blood cardioplegia and all-blood (microplegia), and 3) provide some answers to the question of why do we actually need all of this water from recent data provided by preclinical trials in the research laboratory, and from clinical trials. There is not a whole lot of data gathered over the past 20 years that is relevant to this subject directly, which is a little bit surprising and disappointing. Finally, I will try to use these data to come to some sort of a resolution that you can take home.

Now, as you know, cardioplegia has been the cornerstone of myocardial protection for many, many years. Its most obvious role is to achieve and maintain rapid arrest; the name cardioplegia (2) (cardio = heart; plegia = paralysis) implies this role. Not only is cardioplegia an arresting solution, but it is used as a vehicle to deliver therapeutics to prevent and treat ischemia and reperfusion injuries. Cardioplegia prevents ischemia by 1) lowering the energy demands by arresting the heart, 2) imposing myocardial hypothermia, and 3) delivering various anti-ischemic agents such as adenosine, magnesium, glutamate, and aspartate. Cardioplegia can prevent reperfusion injury not only by limiting ischemia (without ischemia there is no reperfusion injury), but also by controlling the conditions and composition of the reperfusate (3). Specifically, conditions refer to temperature (warm induction to resuscitate the energy-depleted heart, hypothermia during maintenance, warm terminal perfusate, and low delivery pressure to avoid edema and microvascular injury), and composition refers to buffers to limit tissue acidosis, hyperonconicity, agents to reduce calcium, and the addition of therapeutics that directly address the pathophysiology of reperfusion injury (4). The final phase of cardioplegia just before the cross-clamp is released is designed to resuscitate the heart from depolarized arrest and hypothermia (if this is used) in preparation for reanimation and resumption of contractile function. That is a formidable task for a heart that’s been somewhat confused and potentially damaged by ischemia, arrest, hyperkalemia, hypothermia, and reperfusion.

So what is an ideal cardioplegia solution? Arrest is almost universally achieved by hyperkalemia which depolarizes the cell, thereby preventing repolarization and subsequent generation of action potentials. However, hyperkalemia has its darker sides which contribute to postcardioplegia dysfunction and morbidity (5). Efforts have been made to arrest the heart in a polarized state with normal membrane potential by using K$_{ATP}$ channel openers, the combination of adenosine–lidocaine–magnesium (adenocaine), or profound hypocalcemia. Regardless of the modality used to achieve arrest, this arrest presents the surgical team with a quiet, bloodless field, so they can concentrate on the task at hand. Hence, the first role of cardioplegia is to effectively and rapidly arrest the heart. Second, cardioplegia formulations should be based on sound scientific principles developed and tested by appropriately designed and statistically powered preclinical and clinical studies that avoid design flaws such as lack of subject randomization, lack of appropriate treatment blinding, and most of all, insufficient statistical power to declare significance or demonstrate non-significance. A danger in trials claiming non-superiority or equivalence by showing no difference between two or more treatments is that lack of significant difference may represent lack of power to show differences. A Type II error is when statistical differences are missed when actual biological (or treatment) differences exist. Lack of significant difference is often due to insufficient power (e.g., insufficient numbers of subjects or large deviations around the mean) to demonstrate statistical differences, or when the end point has no biological connection or relevance to the process being tested or the actions of the therapeutic. Equivalence does not equal a lack of difference between
treatments in the absence of adequate power. Third, the solution certainly should prevent intraoperative ischemia, and it should resuscitate the myocardium that is presented in an energy-depleted state such as failing hearts or hearts with large evolving infarcts. Fourth, the solution should wash out rapidly upon reperfusion, and reanimation should not be delayed, be sluggish or be irregular once the cardiac motion has started. Finally, the formulation or its mode of delivery should not be inordinately complicated so as to distract the surgical team from the main purpose of the operation—to fix the heart.

Let’s talk a little bit about the history of blood cardioplegia since blood is the focus of the topic at hand. In 1955, Melrose was the first to intentionally arrest the heart with a high-concentration potassium solution (6,7). He used 2 mL of potassium citrate in 20 mL of blood. Therefore, this really is the primordial blood cardioplegic solution if not the first all-blood cardioplegia. However, this formulation was associated with a lot of post-cardioplegia cardiac dysfunction and necrosis (8), and it was subsequently abandoned for almost 20 years. In the interim of the moratorium, other techniques were used, and other arresting agents were used such as acetylcholine, but were not clinically adopted. Other myocardial protection strategies such as aortic cross-clamping to induce ischemic (anoxic) arrest with hypothermia to prevent damage secondary to ischemia were used to still the heart. However, in 1973, Gay and Ebert (9) resurrected the concept of potassium cardioplegia formulation, rationalizing that it was not the potassium per se, or the concept of depolarized arrest that was at fault for previously observed necrosis, but rather the high dose of potassium and/or citrate (10) that was used. Citrate created a very acidic environment that was not conducive to maintaining cell viability. Depolarized arrest induced by hyperkalemia is now arguably the universal approach to arresting the heart electively during cardiac surgery. Dr. Buckberg’s lab in Los Angeles began looking at blood as a vehicle for cardioplegia in 1978 (11–13). The triggering question was “Why are we using all this water in the form of saline or buffers, when the natural fluid for organ perfusion is blood?” That was really the genesis of blood cardioplegia as we know it today. Since then, Dr. Buckberg’s lab and many other labs around the world have looked at the effectiveness of blood cardioplegia, compared it to crystalloid-based or other formulations, and have investigated an endless list of additives and modalities of use (e.g., intermittent vs. continuous delivery, warm induction, relative alkalotic cardioplegia, terminal warm cardioplegia).

So why is blood a better vehicle for cardioplegia? 1) it has a significantly greater oxygen carrying capacity through binding to hemoglobin, compared to the oxygen dissolved in the crystalloid compartment; 2) blood has better rheological properties than crystalloids, i.e., the presence of red cells and the viscosity factor increases perfusion at the microcirculatory level; 3) blood has endogenous substrates that are used for both aerobic metabolism (free fatty acids) and anaerobic metabolism (glucose). If the heart is in an ischemic state, it will use glucose, while if it is in an aerobic state, it will use predominantly free fatty acids. By using blood, we are optimizing an aerobic state, i.e., creating aerobic arrest to minimize damage from myocardial ischemia with a subsequent potential for reperfusion injury; 4) blood has its own inherent buffering capacity contributed by histidine or histidine-containing molecular moieties; 5) blood contains endogenous antioxidants, such as glutathione, peroxidase, and catalase; and finally, 6) blood has endogenous oncotic properties that resist movement of water from intravascular to extravascular compartments, and thereby reduces myocardial tissue edema formation. The endogenous components of blood make it a good vehicle for cardioplegia for the most part. On the other side of the same coin, there is the presence of white blood cells and cytokines, etc., in the blood vehicle that fuel the inflammatory response to ischemia and reperfusion (14), which needs to be attenuated by specific additives.

During the development of hemodiluted blood cardioplegia in the late 1970s, a 4:1 ratio of blood to crystalloid (diluents for drugs) was primarily used, because it accommodated commercially available additives, such as dextrose-5% in water (D5W), tromethamine (THAM), bicarbonate, and citrate-phosphate-dextrose (CPD) (used in blood banking for anticoagulation of stored blood), and reduced sludging thought at that time to impede microvascular distribution of solution. It was convenient, at that time, to make a bag with those drugs diluted in clinically used and approved concentrations. Hemodilution does reduce Rouleau formation, to some extent, at least theoretically, which was another consideration for adopting hemodiluted blood. The modest hemodilution of 4:1 blood cardioplegia maintains essentially all the above advantages of blood, while avoiding the accumulation of a crystalloid load with high-volume usage as would be encountered during long cases and continuous delivery.

Now, as we said earlier, in 1996, Dr. Philippe Menasché, formally brought up the question of why we need all this water contained in hemodiluted blood cardioplegia formulations? Along with Dr. Calafiore, in Italy, Dr. Menasché reported the concept of a minimally hemodiluted blood cardioplegia based on improvement of the rheological properties over that of the hemodiluted blood, and maximizing the oxygen delivery by increasing hemoglobin, by using as few additives as possible, in this case, limited to commercially available formulations of potassium and magnesium—potassium to create depolarized arrest, and magnesium to protect the heart from intracellular calcium influx and overload. This approach was practical, and was simplistic since it limited the complexity of composition, i.e. having
to add different components at different concentrations or a different times, and it was less expensive. The simplicity also stems from a simple syringe-based system linked to a tubing from the oxygenator, which delivered potassium and magnesium. The volume delivery rate could be synchronized with the blood flow rate to get the amount of potassium and magnesium that was desired. Of course, we have more sophisticated delivery systems now, such as the computerized Myocardial Protection System (MPS) by Quest Medical, that can do this more accurately with flexibility to change from crystalloid to blood, and anything in between, and on the fly. Importantly, Dr. Menasché did stipulate that microplegia as envisioned by him was indicated for tepid to warm cardioplegia only. This limitation of its original use narrows the field of uses, but also impacts the basis for testing the microplegia in other circumstances in which other modalities are used, i.e., hypothermia.

**EXPERIMENTAL STUDIES**

So let’s discuss some of the preclinical studies that have investigated the efficacy of miniplegia, as it was called by Dr. Menasché, or microplegia as it is called now, vs. hemodiluted blood cardioplegia to see if there is evidence of superiority of one formulation over the other. In our laboratory, Dan Velez and colleagues (15) performed a study using an anaesthetized open-chest canine model of on-pump revascularization of acute coronary occlusion. The left anterior descending (LAD) coronary artery was occluded for 60 minutes off-pump. Cardiopulmonary bypass was then instituted, and the heart was arrested with 5-minute antegrade dose of either 4:1 blood cardioplegia or microplegia, both delivered by the MPS. Delivery was then converted to retrograde cold continuous cardioplegia for 55 minutes simulating the time period that the LAD (and other clinical target vessels) was bypassed; the LAD ligature was released after 30 minutes to simulate revascularization. A terminal “hot shot” was delivered antegrade before release of the cross-clamp, followed by 30 minutes in the beating empty state, and then 30 minutes in the beating working state after discontinuing cardiopulmonary bypass. Essentially, the protocol mimics an isolated single vessel CABG procedure. What we found was that if you compare the dilute to the all-blood cardioplegia, 1) the crystalloid load was much higher with the dilute cardioplegia as expected, particularly since there was retrograde continuous delivery for such a prolonged period of time. That significantly lowered the average systemic hematocrit compared to the microplegia. 2) the tissue water content of various different organs trended toward being lower with all-blood microplegia, including both the non-ischemic area as well as in the ischemic-reperfused areas. However, tissue water content in the duodenum and in the kidney was significantly lower with microplegia compared to 4:1 blood cardioplegia. 3) infarct size, measured by the vital stain triphenyltetrazolium chloride (TTC) was very similar in both cardioplegia groups: 4) the post-reperfusion LAD endothelium showed dysfunction in the microplegia group, and 5) the number neutrophils that adhere to the LAD coronary vascular endothelium was greater in the microplegia group consistent with observed endothelial dysfunction and an inflammatory response to ischemia-reperfusion: 6) segmental contractile activity (shortening during systole), measured using sonomicrometry crystals placed in the ischemic-reperfused and non-ischemic segments, was only partially restored in the LAD zone of either group, with no appreciable difference between groups. Hence, surgical revascularization does not immediately restore contractile function, and hemodiluted blood cardioplegia or microplegia does not affect the restoration of contractile function. Note also that the differences in coronary artery endothelial dysfunction observed above does not translate into differences in contractile function of the myocardial segment. In summary, microplegia had less crystalloid burden, and decreased systemic hemodilution, and was associated with less tissue edema, but was associated with more neutrophil accumulation and endothelial dysfunction on the ischemic-reperfused coronary artery, but infarct size and loss of segmental contractile activity was comparable between the two groups.

Another study was reported by McCann and colleagues (16), in which anesthetized pigs were placed on arterial-femoral bypass, and the heart was made globally ischemic (normothermic) for 30 minutes. This ischemic period is sufficient to create a model of the energy-depleted or failing heart; clinically these hearts have a high incidence of postcardioplegia failure and mortality. The ischemic hearts were randomized to receive either 4:1 hemodiluted blood cardioplegia or microplegia, both given antegrade on a continuous basis, thereby closely mimicking the model used by Valez et al. (15), but on a global level rather than a regional level, and with continuous antegrade rather than continuous retrograde delivery. The primary end points in this study were echocardiography for left ventricular mass (interestingly not function), and myocardial edema by histology rather than by direct assessment of tissue water content. The authors found the same general observations as Velez et al. i.e., there was more myocardial edema (therefore more mass) with 4:1 hemodiluted blood cardioplegia compared to microplegia. Cardiac function was not measured.

**CLINICAL TRIALS**

There are not a lot of clinical trials specifically testing the efficacy of hemodiluted vs. microplegia. The prospective
trial by Hayashi et al. (17) looked at 4:1 blood cardioplegia vs. microplegia in elective isolated CABG patients. There were 35 participants in each group, so it was a relatively small study with standard formulations. The 4:1 formulation had high potassium concentration, THAM for buffering, CPD to chelate calcium for hypocalcemic reperfusion. Therefore, this reflects to original Buckbergian formulation. This was compared to microplegia with only a high concentration of potassium and magnesium. The strength of this study is that it had a common delivery mode consisting of antegrade induction, continuous retrograde or intermittent antegrade delivery (based on surgeon preference) every 30 minutes for 2 minutes, given at 30°C. By design of the formulation, microplegia was associated with less crystalloid additive to the delivered cardioplegia. Relevant to the clinical outcomes, there was a greater incidence of normal sinus rhythm at the time of reanimation. The peak plasma creatine kinase - myocardial band (ck-MB) fraction was lower, and there was less inotrope use. So there was a demonstrable benefit to microplegia over the 4:1 blood cardioplegia.

El-Hamamsy and colleagues (18), in 2004, also conducted a prospective randomized trial comparing 4:1 hemodiluted blood cardioplegia to “miniplegia”, both used under tepid (30°C) temperature conditions, delivered continuously retrograde. Remember that the original indication for use of miniplegia was under warm or tepid conditions. Again, a low number of patients were enrolled in this study; 25 in the 4:1 blood cardioplegia group, and 34 in the miniplegia group. The patients were low as well as moderate-risk patients scheduled for primary CABG. However, more patients in the miniplegia group had a history of myocardial infarction (MI) or preoperative left ventricular dysfunction (53% vs. 35% in 4:1 group), suggesting that the miniplegia group was actually a higher risk demographic group than the 4:1 blood cardioplegia group. As expected, there was significantly more crystalloid cardioplegia solution given in the 4:1 group vs. the miniplegia group (576 ± 294 vs. 46 ± 14 mL). There were no statistically significant group differences in the incidence of postoperative low cardiac output syndrome, length of hospital stay, and stroke rate. In addition, there was no statistically significant group difference in cardiac troponin T (Tn T) release at 24 hours postoperatively. However, Tn T was unexpectedly higher in the miniplegia group 48 hours postoperatively. The investigators conclude that there were no differences in clinical outcomes between miniplegia and 4:1 blood cardioplegia, but “Minicardioplegia may be the optimal method or myocardial protection because of low cost, ease of use and lack of hemodilutive effect.” However, this conclusion is not entirely supported since neither cost nor ease of use metrics were measured or compared. The statistical power of key end points with this few patients was not reported, so that the basis for equivalence deemed by lack of statistical difference may be a Type II error of no difference detected when there actually may be a biological (clinical) difference.

Another clinical trial by Algarni and colleagues (19) involved prospectively collected, but retrospectively analyzed data from sequentially enrolled patients. The patients in the 8:1 blood cardioplegia group were enrolled between 1998 and 2000, and the patients for the microplegia group were enrolled between 2004 and 2006. The number of patients enrolled were greater than in previously discussed clinical studies—1,980 in each group. The study design incorporated propensity matching which is an accepted method to equalize the level of risk between the groups. Ninety percent of patients received cold antegrade induction and intermittent maintenance every 15 minutes. Continuous retrograde was reserved for the redo and the high-risk patients, so there was a little bit of difference in protocol based on surgeon anticipation of risk category. There was a terminal hotshot before cross-clamp release, and a common delivery system was used in all cases (the Quest Myocardial Protection System (MPS)). The strength of this study is its higher numbers for greater statistical power, and propensity matching to equalize risk factors between groups. The weaknesses include sequential enrollment of patients who were treated with one protocol early on and another protocol later in the sequence, which could have introduced changes in treatment over time. As expected, there was more crystalloid delivered in the hemodiluted group for the same amount of total blood cardioplegia used. There was no significant difference in mortality (1.2%), which is not surprising, since mortality is low in recent years, particularly in these low-risk patients. However, the incidence of low cardiac output syndrome was significantly less in the patient cohort receiving microplegia, and this difference persisted in a subset analysis of elderly patients and high-risk patients. Hence, the investigators conclude superiority of microplegia over hemodiluted blood cardioplegia.

Another clinical trial reported in 2009 by Albacker and colleagues (20), retrospectively reviewed data from 295 consecutive patients older than 75 years in which either microplegia (n = 144) or 4:1 blood cardioplegia (n = 151) was used. The authors found that in-hospital mortality, acute renal failure and confusion were higher in the microplegia group when propensity matching was not used. However, when propensity matching was used after the patients were enrolled and grouped to adjust for differences in pump time, cross-clamp time, procedure type and surgeons, the group differences disappeared. The investigators concluded that microplegia is safe in patients older than 75 years. One must scrutinize the effectiveness of propensity matching used in this manner.
Onorati et al (21) conducted a study comparing 4:1 blood cardioplegia and microplegia enhanced with adenosine-lidocaine-magnesium and insulin (ALMI microplegia). This study, therefore, has an added confounding factor of different additives between groups so that any differences in outcomes cannot be wholly assigned to hemodilution or whole blood. In this study, patients with unstable angina (therefore higher risk patients) were prospectively randomized to 4:1 blood cardioplegia or the ALM microplegia; 40 patients were included in each group. Again, a low number of patients raises the question of the adequacy of statistical power to demonstrate differences in major endpoints. However, as opposed to other studies, the authors demonstrated that statistical power was indeed sufficient in key end points. There was no significant difference in mortality between groups, as we have seen in other studies before, but there was a significantly lower plasma troponin I and lactate sampled from coronary sinus blood in the enhanced microplegia group compared to the 4:1 cardioplegia group. In addition, there was a significantly higher postoperative wall motion score and cardiac index in the enhanced microplegia group compared to the standard 4:1 hemodiluted blood cardioplegia group. In addition, there were fewer blood transfusions and use of packed red blood cells in the ALM microplegia group, even though other blood products were either the same or actually higher in the ALM microplegia group. But importantly, especially for health-care economics, there was a significantly shorter hospital stay in the ALM microplegia group. As stated above, it is not clear from this study whether microplegia per se or the additives ALMI contributed to the advantages in microplegia over hemodiluted blood cardioplegia.

It is often thought that meta-analyses will bring some of these convergent and divergent results together by combining individual studies and thereby increasing the patient numbers and assumedly statistical power. Such a meta-analysis was conducted by Gong et al. (22) very recently in the hemodiluted blood cardioplegia vs. microplegia field. After analyzing 77 studies, Gong et al. (22) distilled a final collection of five studies that fit their criteria of a prospective, randomized primary studies focused on hemodiluted vs. microplegia. The primary outcomes considered were cardioplegia volume, the incidence of low cardiac output syndrome, spontaneous resumption of normal sinus rhythm, and perioperative myocardial infarction (MI). Other outcome variables were not uniformly collected in all studies, so they were omitted in the analysis by Gong et al. (22) This group found that, as expected, there was a significantly lower volume of crystalloid given (not total volume of blood cardioplegia formulation) as part of the microplegia vs. hemodiluted cardioplegia. There was no significant difference in the incidence of low cardiac output syndrome, which they attributed to endothelial dysfunction, consistent with the Valez et al (15) study discussed above. However, there was no data in the included clinical studies to support (or even suggest) this speculation. In addition, there was also no difference in spontaneous reanimation, or perioperative MI. So, instead of clarifying comparisons between hemodiluted and non-diluted blood cardioplegia relative to other studies, the meta-analysis by Gong et al. (22) introduced more inconsistencies. The weaknesses in this type of study include it is a literature review rather than an analysis of the datasets themselves where the individual patient data are coalesced and reanalyzed. But the study by Gong et al. (22) did state that larger prospective randomized clinical trials are needed to test differences in efficacy of hemodiluted and non-diluted blood cardioplegia.

END POINTS IN FUTURE STUDIES

What end points would be discriminative for such a larger clinical trial? We know that mortality is not a good end point because mortality is low in low-risk patients, with little room for improvement. It would be a good end point in studies with high-risk patients since mortality is higher, and efficacy of formulation differences and additives are often expressed in this patient population. Perhaps discriminating end points of choice may be related to morbidity, as in some of the studies discussed above, and may include indices of left ventricular performance, incidence of atrial fibrillation, reanimation with spontaneous sinus rhythm, need for inotropic support in protocol-driven study designs, incidence of neurological or renal damage length of intensive care unit or hospital (again with protocol-driven designs). Importantly, the end points must be linked to the physiological effect of the treatment, or the comparison is superfluous. So I think new studies have to incorporate end points related to morbidity, and go beyond the mortality end point.

SUMMARY AND CONCLUSIONS

To conclude, microplegia has all of the advantages of whole blood, such as 1) superior oxygen carrying capacity, 2) osmotic properties that would reduce tissue and extracellular edema, compliance changes, and left ventricular filling defects that may contribute to postcardioplegia contractile dysfunction (stunning), 3) endogenous buffers, and 4) simple and cost-effective formulation compared to more complex formulations with multiple additives. The 20% or less hemodilution in the hemodiluted formulation should not drastically reduce these attributes except in long cases using continuous delivery. In such cases, systemic hemodilution can be avoided using microplegia (23). The original indication for use advocated by Dr. Menasché was for tepid and continuous delivery. Now clinical use is
reaching into hypothermic delivery and non-continuous (intermittent) delivery without evidence to base the new use. Does microplegia have the same efficacy in those clinical environments? The preclinical data indicate superiority of microplegia over hemodiluted blood cardioplegia in end point variables other than postcardioplegia cardiac functional variables, but are unclear on postcardioplegia cardiac function. The clinical studies suggest there is some benefit on postcardioplegia functional end points, but a clear picture of outcomes is not presented because the studies are largely not prospective, randomized, and blinded with sufficient statistical power. In addition, the influence of delivery modality (continuous vs. intermittent) or temperature management (hypothermia vs. tepid), and the efficacy in patient subpopulations are unknowns, and require stepwise, logistical trials. These are questions that will provide much fruitful research in the coming years. However, funding for these studies is a major problem, and the lack of funding would impede progress in this area. National Institute of Health funding is highly competitive, and programmatic funding for a head-to-head trial of hemodiluted vs. all-blood cardioplegia as needed will be hard to obtain. Likewise, corporate funding for such a non-product trial is difficult to obtain. Piggy-backing additives in hemodiluted and all-blood environments would be one way to approach this issue, but it would require more extensive and more complex (and more expensive) studies.

Just a final hopeful note: The baby need not be thrown out with the bathwater in adopting an all-blood formulation. That is, the additives used in hemodiluted formula-tions, tested for over 30 years, and to which the surgical in-duction. 

REFERENCES