REVIEW

Ex Vivo Lung Perfusion: A Review of Current and Future Application in Lung Transplantation

Kareem Ahmad · Jennifer L. Pluhacek · A. Whitney Brown

ABSTRACT

The number of waitlisted lung transplant candidates exceeds the availability of donor organs. Barriers to utilization of donor lungs include suboptimal lung allograft function, long ischemic times due to geographical distance between donor and recipient, and a wide array of other logistical and medical challenges. Ex vivo lung perfusion (EVLP) is a modality that allows donor lungs to be evaluated in a closed circuit outside of the body and extends lung donor assessment prior to final acceptance for transplantation. EVLP was first utilized successfully in 2001 in Lund, Sweden. Since its initial use, EVLP has facilitated hundreds of lung transplants that would not have otherwise happened. EVLP technology continues to evolve and improve, and currently there are multiple commercially available systems, and more under investigation worldwide. Although barriers to universal utilization of EVLP exist, the possibility for more widespread adaptation of this technology abounds. Not only does EVLP have diagnostic capabilities as an organ monitoring device but also the therapeutic potential to improve lung allograft quality when specific issues are encountered. Expanded treatment potential includes the use of immunomodulatory treatment to reduce primary graft dysfunction, as well as targeted antimicrobial therapy to treat infection. In this review, we will highlight the historical development, the current state of utilization/capability, and the future promise of this technology.

Keywords: Ex vivo lung transplant; EVLP; Donor lung utilization; Donor lung conditioning; Cost-effectiveness; Lung transplant outcomes; Lung transplant volume

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Key Summary Points

Ex vivo lung perfusion (EVLP) is a rapidly developing and increasingly utilized technology to increase the lung donor pool for transplantation.

EVLP has evolved into a device for logistical optimization of the lung transplant process and increases the tolerability of cold ischemic time through its use.

Monitoring of the lung(s) on EVLP allows for more thorough evaluation of the lung(s) and increases confidence in donor lung utilization.

Issues such as atelectasis and pulmonary edema may be improved through the use of EVLP. Future opportunities for treatment of donor lung infection and immunomodulation exist on the EVLP platform.

Rapid and more universal adoption of EVLP remains limited by resource utilization.

DIGITAL FEATURES

This article is published with digital features, including a video, to facilitate understanding of the article. To view digital features for this article go to https://doi.org/10.6084/m9.figshare.19189520.

INTRODUCTION

Lung transplantation is the ultimate treatment for patients with various forms of end-stage lung disease. However, a large discrepancy between the number of patients awaiting transplantation and the number of suitable donor lungs has always existed. Despite a mismatch in supply and demand, utilization rates of donor lungs remain around 20% [1]. Often donor lungs are discarded over concerns regarding quality, time, or logistical constraints from organ procurement to transplantation, and the potential for future graft dysfunction [2]. This scarcity of appropriate donor lungs leads to a mortality rate of up to 30% of wait-listed patients [3]. Opportunities to increase the number of donor lungs available include the use of extended donor criteria (ECD) and donation after circulatory death (DCD) [4]. Ex vivo lung perfusion (EVLP) is a technology to optimize allograft assessment and function and in these less conventional situations to expand the donor pool for patients on the waiting list.

EVLP is a modality that allows donor lungs to be evaluated in a closed circuit outside of the body and extends lung donor assessment prior to final acceptance for transplantation [5]. Three EVLP protocols are currently in use worldwide emerging from Lund, Toronto, and the Organ Care System Lung [6]. The Lund protocol is the original protocol adapted further into the Toronto protocol, which is now the most commonly used. The third protocol includes the use of a transportable device that uniquely eliminates the need to keep lungs in static cold storage prior to transplant and effectively reduces the cold ischemic time [7]. There are currently four commercially available EVLP devices using one or a hybrid of the three standard protocols: Lung Assist™ by Organ Assist®, XVIVO Perfusion System (XPS)™ by XVIVO®, Vivoline LS1™ by Vivoline Medical®, and OCS™ by TransMedics® [9–11] (Fig. 1).

Each EVLP system consists of perfusion and ventilation in an enclosed and controlled unit to monitor function and maintain asepsis of the lungs. Lungs are ventilated through an endo-tracheal tube. A vascular circuit consists of outflow from the left atrium through a reservoir, pump, oxygenator, heater-cooler unit, and a leukocyte filter before entering back through the pulmonary artery [8]. The protocols can vary in perfusate, rate of flow, target left atrial pressure, and ventilatory settings [6].

While the lungs are on the circuit, a variety of assessments can be performed to evaluate organ quality. Depending on the system, this can include gross physical examination,

radiographic imaging, bronchoscopy/airway inspection, pressure measurements to include lung compliance, and gas exchange. These dynamic assessments allow the treating team to monitor complications such as pulmonary edema, atelectasis, as well as identify suspicious tumors or mechanical issues such as iatrogenic procurement injury that may ultimately compromise the graft's successful transplantation [12]. EVLP, in some cases, provides a mechanism to improve suboptimal donor lung quality through mechanical recruitment of atelectatic segments, therapeutic aspiration of secretions via bronchoscopy, and intravascular administration of anti-inflammatory and antibiotic therapies [13]. These benefits may significantly increase the number of lungs suitable for transplantation, thereby contributing to increased transplant volume. Though yet to be shown, the rehabilitation and utilization of even a small percentage of suboptimal donor lungs deemed otherwise unacceptable for transplant, may result in reduced waitlist mortality.

In this review, we present a brief overview of EVLP followed by an analysis of advantages and disadvantages in the current lung transplant landscape. We will then look to the future and evaluate potential uses for EVLP moving forward. This article is based on previously conducted studies and does not contain any new studies with human participants or animals performed by any of the authors. The lung transplant recipient involved in the case study provided consent for the details of his clinical course to be disclosed in this manuscript.

HISTORY OF EVLP

EVLP has been tested in a myriad of animal models including mice, guinea pigs, rabbits,
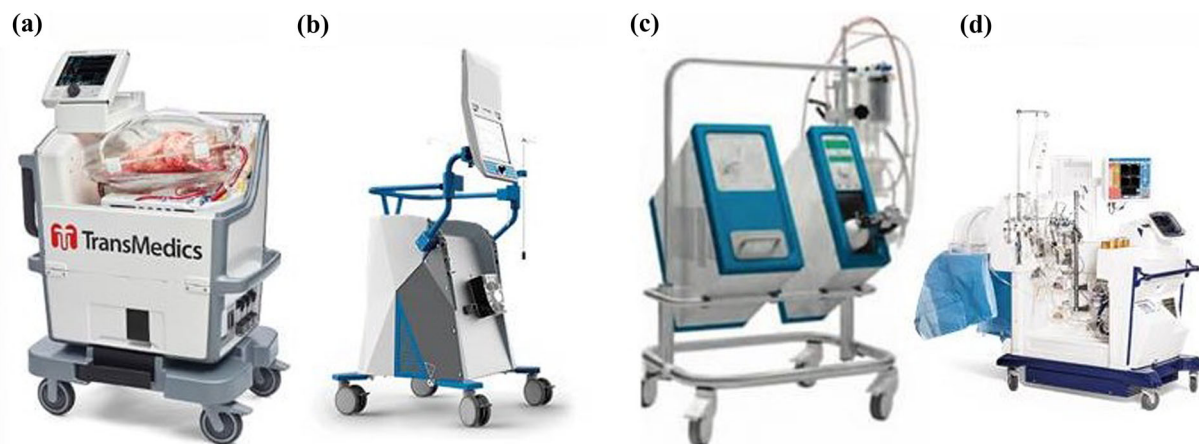
System	Characteristics
(A) OCS* (Transmedics, Andover, MA, USA)	Reduced total ischemic time with utilization of recipient blood-based perfusion immediately from time of explantation to transplant.
(B) XVIVO LS (XVIVO Perfusion AB, Göteborg, Sweden)	Smaller device with easy to use interface and disposable single organ set for reduced setup. Device is no longer available for sell.
(C) Lung Assist (Organ Assist, Groningen, Netherlands)	Isolated perfusion device of organ in- and ex-situ for evaluation in hypothermic and normothermic conditions. Compatible for use with any pre-existing ventilators.
(D) XVIVO XPS* (XVIVO Perfusion AB, Göteborg, Sweden)	Unique organ chamber is designed for facilitation of radiographic imaging. Large device with full perfusion, ventilation, and monitoring requiring dedicated space.
	*FDA approved

Fig. 1 Commercially available EVLP systems

dogs, and pigs as early as 1970 [14]. While animal models initially served as a cost-effective means for "proof-of-concept" pre-clinical model of EVLP, small organ size limited translatability to human use [15]. Given porcine lungs are similar to human lungs in size, physiology, and gene sequence making, it has become the preferred animal model [16]. The close relationship between porcine and human lungs allowed researchers to test the same equipment for the EVLP circuit that went on to be used in human clinical trials [15].

The first clinical application of EVLP in humans took place in 2001 at the University Hospital of Lund, Sweden. Steen and his colleagues originally intended for EVLP to be used as an assessment tool for lungs procured from a DCD donor [17]. During this first attempt, lungs were on the circuit for a brief time of 65 min but allowed for bronchoscopy and radiography to be completed [18]. Extending their success, the Lund group went on to show utility of EVLP in improving suboptimal/marginal lungs and completed the first transplant of lungs not meeting traditional donor criteria (ECD) in 2005 [19, 20].

The Toronto Lung Transplant Program took EVLP a step further by allowing an extended perfusion time of up to 12 h in a pre-clinical model [21]. This now meant that not only could lungs undergoing EVLP be evaluated closely over a longer time period, but also potentially reconditioned prior to transplantation. Building on the success of this technique, the initial clinical trial data reported the successful transplantation of 20 out of 23 marginal lungs after a period of 4 h of perfusion with improved gas exchange [22]. The study primarily highlighted that the use of EVLP overcame the historically significant higher rate of primary graft dysfunction (PGD) in marginal lungs in comparison to conventional donor lungs. In fact, there was a tendency of reduced rates of PGD at 72 h after transplant in the EVLP group. After successful use of EVLP by the Lund and Toronto groups, other centers worldwide began to experiment with their own EVLP circuits, although many lungs at these centers ultimately were not utilized [23]. However, use of the Lund and Toronto protocols over time

increased conversion rates of utilization up to 90% for marginal lungs [13].

Advancements in technology have made EVLP more accessible and efficient since initial use (Fig. 2). The first models of EVLP in Lund and Toronto were operated manually and were performed in Plexiglass boxes [24]. As stated above, there are now four commercialized semi-automatic devices available each based on the original protocols, or in one case, a blend of them (Fig. 1). A major milestone in EVLP was the successful development of the first portable EVLP system, The Organ Care System^{TM} (OCS^{TM}) Lung, a blending of Lund and Toronto protocols, eliminating the need for static cold preservation during transportation from the donor directly to a recipient. Results of the OCS^{TM} Lung clinical trial (INSPIRE) were reported in 2018 [25]. Uniquely, recipient derived blood is used during the OCS^{TM} EVLP process. Physiologic donor lung preservation with the OCS Lung was shown to be non-inferior to conventional lung transplant, with no significant difference in PGD, 30-day mortality, and 1-year survival. While this advancement revealed the safety and effectiveness of sparing prolonged cold ischemia, the direct effect of a portable versus static EVLP system remains unknown. Of note, a subsequent study of the OCS^{TM} Lung showed nearly 87% conversion rate of ECD lungs [26].

As highlighted above, the goal of EVLP has been to increase the number of viable donor lungs and ultimately, a yet to be seen, decrease in mortality on the lung transplant waitlist. EVLP initially provided a platform for evaluation of the lungs for a short period of time after DCD to assess their viability for transplant. As techniques have improved and technology evolved, it has become possible to perfuse lungs ex vivo for longer periods of time to objectively assess lung quality and intervene to improve organ function. As we will detail later, EVLP can be further expected to evolve into an organ "repair" tool [27] (Fig. 2).
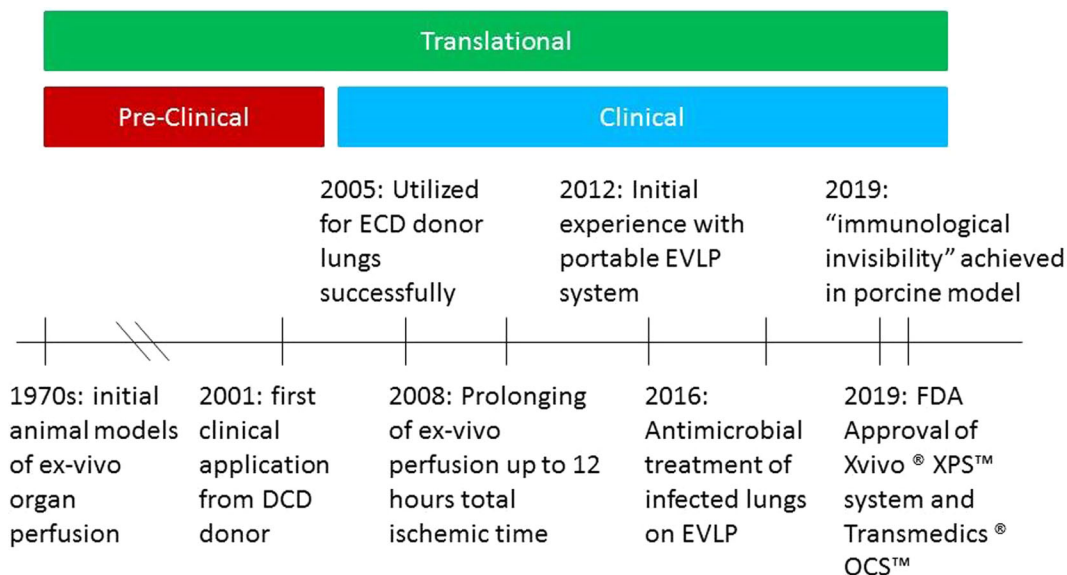
Fig. 2 Major developments in timeline of EVLP

APPLICATION OF EVLP IN LUNG TRANSPLANT

The practical application of EVLP is best understood within the context of a standard lung transplant. There is heterogeneity in the organ allocation process around the world. In the United States, organ allocation is done by the organ procurement organization (OPO) local to the donor hospital. Each OPO has its criteria for evaluation and donor management which requires juggling the needs of multiple organ systems. Lungs are generally allocated within a certain geographical radius by blood type, height range, and lung allocation (patient acuity) score. Should an organ meet a recipient center's requirements (detailed below), lung(s) are accepted pending direct visual inspection at the time of procurement. When available, the lungs are evaluated by chest imaging, bronchoscopy, and serial blood gas analyses prior to procurement. Gross visual evaluation is done after the opening of the thoracic cage. Sometimes, physiologic testing of individual segments of the lung can be done by regional blood gas analysis at time of procurement. Upon acceptance in the operating room, the lungs are deflated and dissected away from

the central airways and heart. This is followed by antegrade perfusion by a cooled acellular solution to help rapidly halt metabolism and clear donor blood which may contain damaging agents. Often corticosteroids and intravenous vasodilators (nitroglycerin/prostanoids) are used to help preserve vascular function and limit the inflammatory response to ischemia. Retrograde perfusion is done to exclude and remove pulmonary thrombi. The organ is preserved in the acellular, cooled solution for rapid transport to the center where a recipient awaits. After a recipient's native lungs have been explanted, the donor organ is anastomosed, reperfused, and rewarmed by the recipient's circulating blood. After adequate gas exchange and hemostasis is achieved, the thoracic cavity is closed and the patient is returned to the intensive care unit for monitoring [28].

As we will show, EVLP can be used to overcome some of the key logistical challenges involved in the conventional lung transplant process (Fig. 3).

DONOR QUALITY

Traditional criteria for lung donors originated in the 1980s [29]. Based on initial criteria, the

ideal donor is brain dead with preserved cardiovascular function, young (under age 55), with no acute or chronic lung disease, without significant smoking history, clear radiographically and on bronchoscopy, and geographically close to the recipient. To expand the donor pool, these criteria have been extended to include donors who may meet limited standards. To mitigate the risks of suboptimal lung allograft function in these cases, a lung donor scoring tool to predict outcomes and utilization rate was developed to confer confidence in transplanting these less optimal organs [29, 30]. Likewise, lungs from DCD donors can be successfully utilized, although there are unique challenges such as the unpredictability of time from withdrawal of life support to death (agonal time) as well as the occurrence of atelectasis and de-recruiting after withdrawal [31]. In addition, there may be limited data (bronchoscopy in particular) available prior to the time of procurement for DCDs given restrictions on invasive procedures for non-brain-dead donors in

certain situations. These challenges can lead to "dry runs" in which an organ is evaluated in the operating room and deemed not transplantable due to excessive agonal time or poor organ quality [32]. "Dry runs" are resource intense, diverting surgical teams and transport systems away from other healthcare system needs. Retrospective analysis of referring Belgian centers showed that up to 7.4% of lung donors were declined at time of procurement due to concerns about organ quality [33].

ISCHEMIC TIME

The duration of time between procurement and implantation into the recipient is defined as the ischemic time—without perfusion. It consists of one or two phases, "cold" $\pm$ "warm" ischemia. Traditionally, an organ will only undergo "cold" ischemia during which preservation is targeted by reduction in metabolism using temperature regulation, using 4 °C solution and storage in
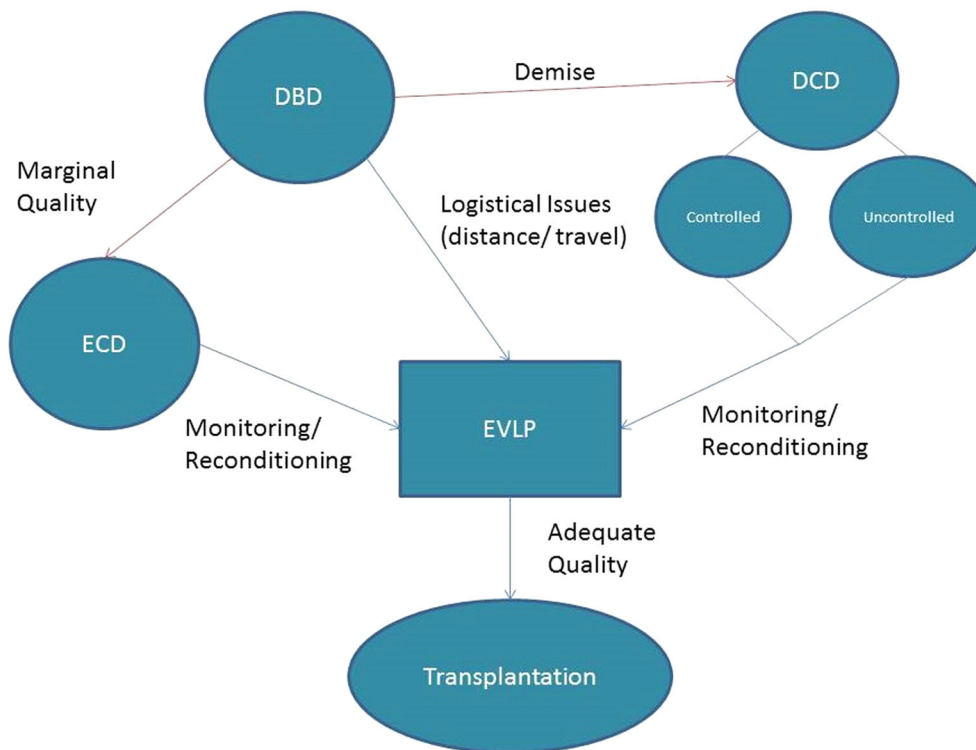

Fig. 3 Utility of EVLP depending on donor type

an ice cooler. This is ideally limited to 4–6 h to ensure adequate function of the lung allograft, though studies suggest up to 9–10 h can be acceptable [34]. Travel distance is limited by cold ischemic time. Normothermic perfusion phase (also called warm ischemic time) describes the duration during which the organ is perfused and ventilated at physiologic temperature, 34–37 °C outside of the body. With the addition of a normothermic perfusion phase, total preservation time can be significantly extended [35].

LOGISTICAL ISSUES

Transplantation is a complex workflow that involves a myriad of healthcare providers and logistical support personnel. Organ procurement organizations (OPO) are regional and coordinate the recovery of multiple organs by distinct transplant teams. Organ procurement teams travel to the donor, the procured organ then is transported from the donor center to the recipient hospital, and inclement weather may result in unsafe travel at any point along the timeline. In most cases, while procurement is occurring, the recipient is undergoing preparation, and in some cases, explantation of the native lungs to minimize ischemic time. Many lung transplant recipients are admitted from home at the time of donor organ availability, thus travel to the transplant hospital and pre-operative preparation may delay the ability to begin the transplant. The need to optimally coordinate and time these processes ultimately affects the success of the transplant. Notable recent data from Australia highlights that up to 26% of all declined donor offers were due to logistical issues [36].

CONVENTIONAL LUNG TRANSPLANT VS. LUNG TRANSPLANT WITH THE USE OF EVLP

Most commonly, cold preservation of the organ begins immediately after organ recovery for transport to the recipient hospital. When EVLP is utilized as an intermediate step, the lungs are either (1) placed on a portable EVLP device without cold preservation (OCS^{TM}) or placed in cold preservation and then (2) transported to a third-party EVLP facility, or (3) taken to the recipient transplant center and placed on EVLP in an operating room near the recipient. Notably, if a third-party facility is involved, the organ is cooled a second time after the completion of EVLP for transport to the recipient's facility for transplantation (Fig. 4).

With use of the portable EVLP device (OCS^{TM}), the donor lungs are not placed on ice, rather directly placed on an ex vivo device to maintain warm perfusion with blood and ventilation. This mobile device is used to transport an organ from the donor center directly to the recipient hospital [7]. At time of removal from the circuit, the lungs are flushed with and immersed in cold acellular solution to allow for time for re-anastomosis (Fig. 4B).

Generally uniform across the platforms, organs are deemed acceptable for transplantation if compliance, airway pressures, and vascular resistance are stable or improving. As prescribed by the Food and Drug Administration for both currently approved systems, the lung(s) must have adequate gas exchange via perfusate gas sampling. The XPS system uses a change in absolute oxygen tension between the arterial and venous access during ventilation with 100% oxygen and a positive end expiratory pressure (PEEP) of 5 cm H20, with two values greater than 350 mmHg to deem an organ acceptable [37]. For the OCS^{TM} Lung, a partial pressure of oxygen (PaO2) to fraction of inspired oxygen (FiO$_2$) (P/F) ratio over 300 mmHg while ventilated using a PEEP of 5 cm H$_2$O and FiO$_2$ of 21% (room air) is required [38]. Radiographic improvement is desirable with recruitment of atelectasis and reduction of pulmonary edema. Lastly, a "collapse" or "deflation" test is performed to ensure normal global deflation (atelectasis) upon disconnection from positive pressure ventilation [22, 25] (See video 1 in the online/HTML version of the manuscript). Recent development of the COMPLETE score and utilization of P/F ratio with inflammatory score (by serial interleukin-6 and -8 measurement) seek to further standardize acceptance criteria across platforms [39].
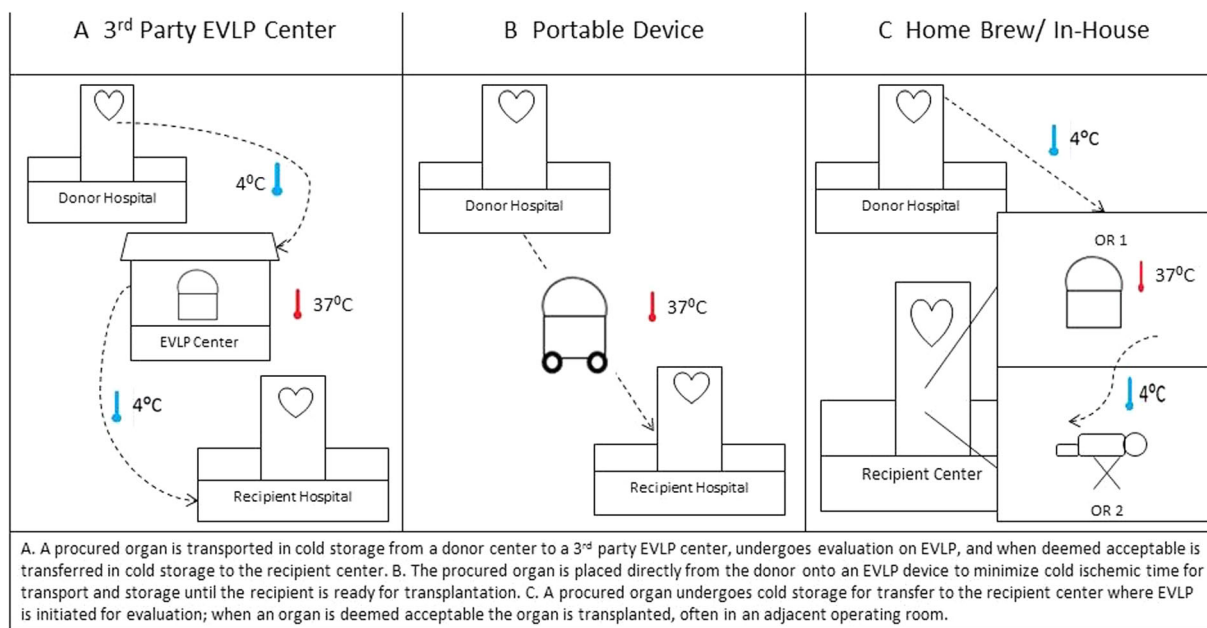
Fig. 4 Schematic of application of EVLP during lung transplant

KEY BARRIERS TO THE USE OF EVLP

The incorporation of EVLP into a lung transplant center's practice involves an initial investment of time and resources. Fundamentally, the components can be conceptualized in three broad categories: personnel cost, the system (i.e., equipment and supplies) itself, and physical space. Depending on the EVLP system selected, there may be a significant initial capital investment for the equipment itself as well as hands-on training for responsible staff members. Estimated initial cost for commercially available systems range from $10,000 up to approximately $250,000, excluding replacement costs of disposable parts [40]. Additionally, operating room space for an EVLP procedure is necessary if a portable system is not utilized. This involves high level institutional commitment to growth of the lung transplant program [41]. Often, a sporadically used system that is resource intensive is difficult to justify in the hierarchy of institutional needs. For institutions less willing or able to make a large initial capital investment, outsourcing to a dedicated EVLP facility may be more practical (Table 1). In

some cases, a center may decide that a combination approach may offer the most flexibility for their program.

Training of transplant staff members on the selected EVLP system(s), guidelines for donor lung referral, logistics of referral, criteria for acceptance of lung(s) from EVLP, impact on timing of transplant cases, and modification of existing procedures and standards of practice are not standardized. The cost effectiveness of integration of EVLP into transplant center practice is an area of ongoing study [42–44]. EVLP has been shown to be profitable at the institutional level through increased transplant program volume, but each individual EVLP case costs approximately $US 40,000–50,000 more than a traditional transplant. The broader role within healthcare delivery on a state or national level remain difficult to determine. The National Health System in the United Kingdom attempted to assess this question during the DEVELOP-UK trial, showing that cost, on average, was about 35,000 GBP more per patient but the trial was stopped early due to a high need for ECMO in the EVLP arm [45].

Table 1 Comparison of modalities

EVLP modality	Initial capital investment	Cost per case	Space requirements	Transplant center staff requirements	Advantages	Disadvantages
Portable system	Significant	Supplies for case, staff pay including travel	Storage when not in use, transport space	Perfusionist(s) travel to procurements	No cold ischemic time, autonomy, may be more cost effective for high volume centers	Requires additional staff member training, availability, and travel to procurement
Transplant center system	Significant	Supplies for case, staff pay	Storage when not in use, dedicated operating room when in use	Perfusionist(s) operate device at institution	Autonomy, may be more cost effective for high volume centers, may allow for research protocols	Requires staff member training, availability, and operating room space
EVLP facility	None	Negotiated bundled fee per case	None	None	Less up-front cost and commitment, may appeal to small to medium-sized programs	Adds additional travel segment to EVLP facility, adds second cold ischemic time to travel to lung transplant center

INCREASING LUNG TRANSPLANT VOLUME

Utilization of EVLP can result in augmentation of overall transplant volume and lung utilization rate, both directly and indirectly. Direct growth can be quantified easily by conversion rate of donor lungs that do not meet conventional center criteria for acceptance, but undergo EVLP, and are subsequently accepted as referenced previously. Indirect growth may also result because access to EVLP can alter patterns of donor offer acceptance and willingness to travel to evaluate donor lungs in person given access to EVLP as a back-up modality for suboptimal quality. Even when EVLP is not actually utilized for a case, there is likely behavior change and more aggressive selection of donor lungs because of its availability. Multiple centers throughout the world have reported significant increases in utilization of donor organs after integration of EVLP, up 70% after a decade of experience [41–43, 46, 47]. More aggressive lung donor utilization, in many cases, may translate to decreased waitlist times, though this may be difficult to demonstrate until there is more

universal adoption of EVLP [48]. The ability of EVLP to increase the pool of donor lungs remains particularly important at the extremes of the spectrum for recipients, those with low waitlist urgency and those with high waitlist urgency.

In addition to its utility in cases of suboptimal lung allograft quality, EVLP allows for the gift of flexibility during a rather strict timeline. As highlighted previously, organizational constraints imposed by multiple factors can lead to viable organs being declined. The additional time afforded by EVLP can be helpful in a situation where procurement occurs in the absence of a comprehensive evaluation. For example, when bronchoscopy or computed tomography (CT) imaging are not possible prior to acceptance of the DCD donor lungs, this which could lead to discomfort accepting the lungs for direct transplantation without ELVP evaluation [49]. One notable example includes the use of EVLP for to permit adequate time for histologic evaluation of extra-pulmonary malignancy [50].

As stated previously, "dry runs" or last-minute donor lung declines can occur even in the operating room. The availability of EVLP allows other lung transplant programs the opportunity to independently evaluate the lungs for their recipients using their own acceptance criteria [36]. Additionally, broader sharing of organs over larger geographic areas and extended travel times are becoming more common, with some programs extending travel distance to up to 5000 nautical miles [51]. Likewise, sometimes there are recipient issues that can lead to extended ischemic times, such as time to arrival to transplant hospital, clinical instability, need for prospective crossmatch, or technically difficult removal of native lungs. One example, the Foch Hospital (Suresnes, Franc) successfully utilized EVLP to offer three hyperimmunized candidates transplantation after providing adequate time for a prospective crossmatch to be completed [52]. Lastly, operating room or surgical team availability may be limited in some situations where there are multiple competing procedures occurring concurrently. Ultimately, extended warm perfusion has been performed using multiple EVLP systems, and pre-clinical data suggest that EVLP may allow for a

'resetting' of adverse physiologic changes that may occur during cold ischemic time, buying time to overcome potential issues as described above [34].

UTILIZATION OF EVLP IN THE UNITED STATES

With increasing availability and familiarity with the use of EVLP for lung transplant, utilization has increased some over the past few years in the United States (Fig. 5). Recent inquiry of data from the United Network for Organ Sharing/ Organ Procurement and Transplant Network shows the number of organs utilizing EVLP range between 4.7 and 6.5% [53]. As more centers gain access to EVLP effectively increasing the lung donor pool, we can anticipate a rise in lung utilization rate which may be accelerated by standardization of protocols and service development [54].

FUTURE UTILITY OF EVLP

Much of the current utility of EVLP has been focused on short-term outcomes such as PGD and 30-days post-transplant mortality. Data from individual institutional retrospective studies and post hoc analyses of clinical trials provide evidence of equivalent 12-month allograft and patient survival of after EVLP versus conventional transplantation [13]. However, the utility of EVLP can extend beyond organ evaluation and extension of ischemic time to incorporate more advanced diagnostic and therapeutic modalities.

Allograft Assessment

An area of great growth and potential within EVLP is the capacity for comprehensive, multidimensional organ monitoring. Gross inspection, such as by weight, can be predictive of suitability [55]. Additionally, sampling by bronchoscopy may play a potential role in future diagnostic testing, even on a molecular level [27]. Beyond conventional chest radiography, broader imaging modalities such as
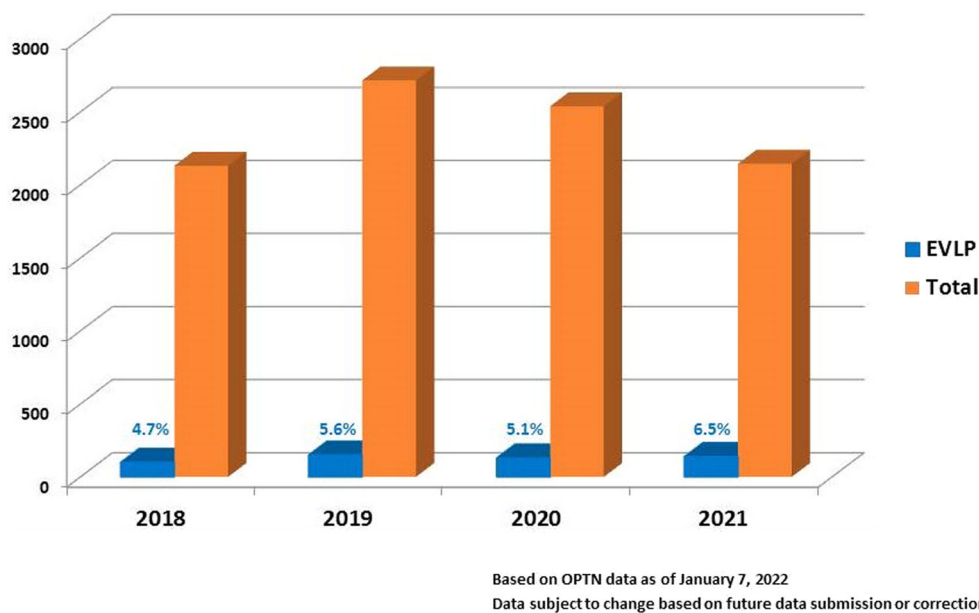
Fig. 5 Deceased donor lung transplants performed in the US from 03/2018 to 10/2021 stratified by use of EVLP prior to transplant

Antimicrobial Treatment

magnetic resonance imaging and ultrasound techniques are being investigated [56, 57]. Perfusate sample assessment in pre-clinical models has identified extracellular biomarker presence and pro-inflammatory gene expression. Certain monitoring modalities, once validated, could help assess organ response to ischemia, quantify inflammation, and help predict future reperfusion injury and/or alloimmune response [58]. However, feasibility of these types of assessment in the clinical setting still requires further study. As a clearer understanding of normal allograft response to EVLP emerges, so too will the potential for interventions to optimize lung allograft function [59, 60].

Antimicrobial Treatment

Donor transmission of infection through transplantation is an inherent risk. Prolonged mechanical ventilation, terminal extubation with DCD, and reduced or absent airway reflexes (to include cough clearance) increase the risk of infection of the donor organ. Infection may result in direct cellular injury and reduced gas exchange, as well as propagation of infection and transmission to recipient during the transplant process [61, 62]. Activation of the recipient immune system by harbored organisms can also lead to reduced immune tolerance [63]. Prior to transplant, donor management involves a risk/benefit ratio for any donor intervention to maximize multi-organ donation potential. Antibiotics can have deleterious side effects, such as aminoglycosides with significant risk of precipitating acute kidney injury. Inversely, localized infection or systemic sepsis can adversely affect donor lung quality [64]. EVLP allows for targeted treatment of donor lungs in isolation, including removal from a toxic milieu of endotoxins. Additionally, it can allow for supra-therapeutic drug levels without associated toxicity to other donor or recipient organs [65, 66]. For example, administration of vancomycin is considered therapeutic if a pre-dose serum level of 10 mg/l can be maintained, however peak levels above 30 mg/l increase the risk of nephrotoxicity. Administered during EVLP, a single dose of 15 mg/kg of donor body weight could consistently maintain a perfusate concentration of 300–400 mg/l for up to 12 h, a level significantly higher than the threshold for

renal toxicity and allowing maximal efficacy of the time-dependent bactericidal effect of this medication. Broad-spectrum coverage with antibacterial agents during EVLP can reduce bacterial load, endotoxin, and alter pro-inflammatory markers with a significant improvement in gas exchange and pulmonary mechanics. Interestingly, inhaled nitric oxide has been shown to have antimicrobial effects, and has been safely used in a pre-clinical model of EVLP for a continuous 12 h at 200 parts per million without associated risk of side effects such as systemic hypotension and methemoglobinemia [67]. Apart from direct transmission of respiratory infection, blood-borne pathogens also pose a threat to recipients. Viral load reduction of hepatitis C with ultraviolet light application during EVLP, specifically with wavelengths of 200–280 manometers (ultraviolet-C) appears to be useful for disinfection and could serve as a method to reduce donor transmission of hepatitis C [68]. Similar reduction of other viruses to reduce transmission such as hepatitis B and human immunodeficiency virus may also prove feasible and effective, thus leading to increased donor lung utilization.
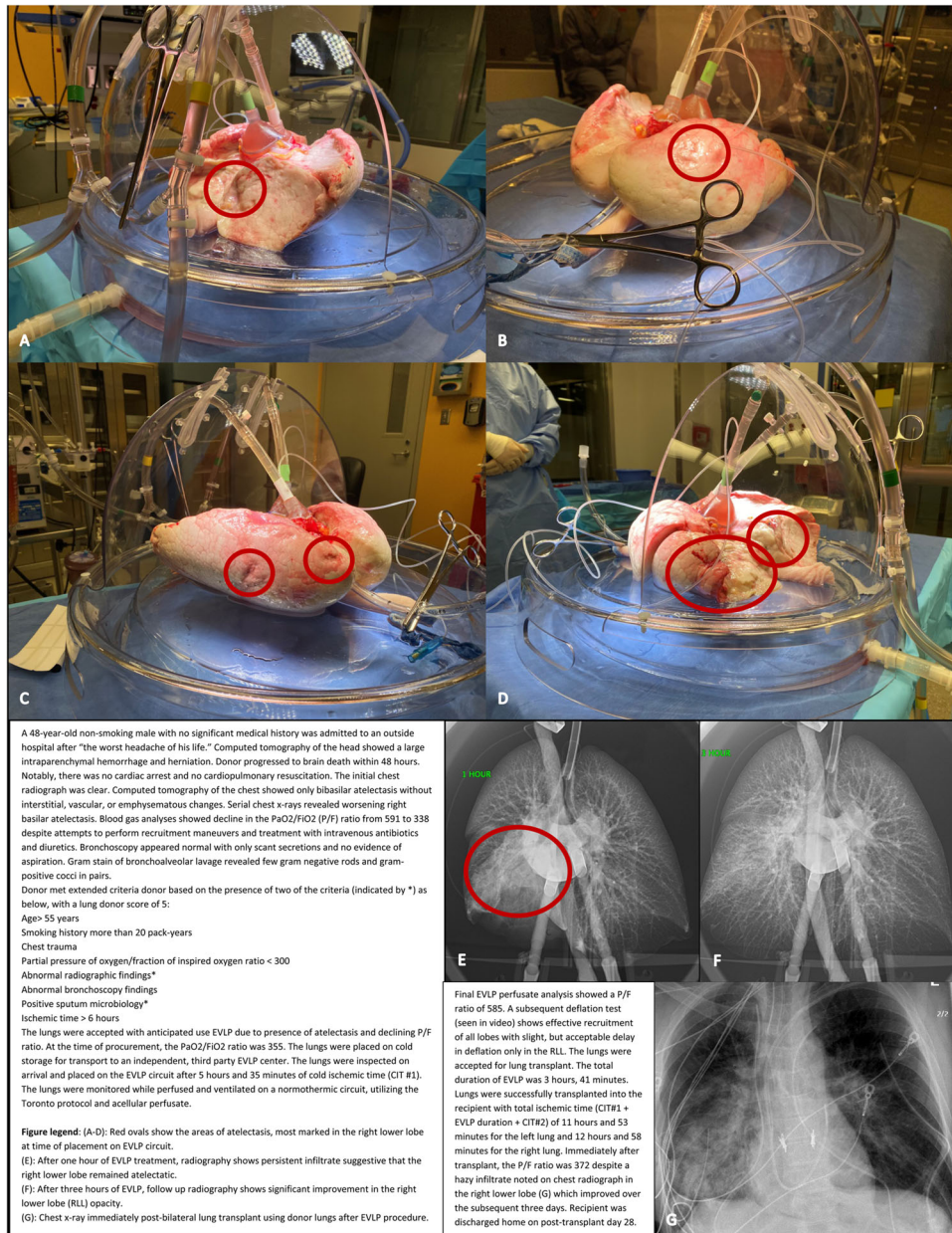
Optimization of Allograft Function

Ultimately, long-term allograft function and recipient survival is the goal of a successful lung transplant. EVLP promises to be an effective platform for directed therapies for immunomodulation as well. Therapies such as anti-inflammatory alpha 1-antitrypsin administration or allograft B-cell depleting rituximab are investigational at present but may eventually be adopted into broader use [69, 70].

Acute and chronic rejection stem from recipient immune system recognition of a foreign organ and are mitigated using systemic immunosuppression [71]. The pro-inflammatory state immediately after transplant can be associated with long-term rejection [72]. Multidrug regimens for immunosuppression, however, increase risk of infection and secondary effects can lead to other end organ damage. Though limited, pre-clinical models of isolated treatment during EVLP with cyclosporine and

methylprednisolone have shown benefit on physiologic parameters and gas exchange during early graft function [73, 74]. Longer-term data remain to be seen. Vector therapy using modified adenovirus/lentivirus could be utilized to deliver gene therapy directly to the organ to reduce pro-inflammatory signaling without exposing other recipient organ systems [75, 76]. Other targeted immunomodulation, RNA based therapies, have been utilized and can selectively be administered during EVLP [77]. Immunomodulation of the donor lungs during EVLP could reduce recognition and sensitization of the recipient immune system and/or increase tolerance markers resulting in less acute and chronic rejection potentially with lower levels of long-term immunosuppression.

CONCLUSIONS

Although use of ex vivo lung perfusion remains in its early phase, multiple studies have demonstrated its feasibility and utility. Increasing use of commercially available and investigational EVLP systems demonstrates the potential of EVLP to further revolutionize lung transplantation. With the passage of time and ongoing investigation, additional advances and potential benefits of EVLP are assured. In the short term, EVLP represents a viable pathway to increase the lung donor pool and potentially reduce transplant waitlist time and mortality. However, this technology holds promise to modulate long-term post-lung transplant outcomes as well. The optimal EVLP system(s) for a particular transplant center depends on individual center practices and resources, thus the availability of distinct EVLP models is critical. Expanded capacity for diagnostic testing to guide lung allograft acceptance and transplantation holds great promise, but perhaps most exciting for the future of EVLP is the emergence of therapeutic options for infection control and immunomodulation. With continued evolution and expanded utility, ex vivo lung perfusion may provide the major leap needed to improve and extend allograft function in lung transplantation.

A 48-year-old non-smoking male with no significant medical history was admitted to an outside hospital after "the worst headache of his life." Computed tomography of the head showed a large intraparenchymal hemorrhage and herniation. Donor progressed to brain death within 48 hours. Notably, there was no cardiac arrest and no cardiopulmonary resuscitation. The initial chest radiograph was clear. Computed tomography of the chest showed only bibasilar atelectasis without interstitial, vascular, or emphysematous changes. Serial chest x-rays revealed worsening right basilar atelectasis. Blood gas analyses showed decline in the PaO2/FiO2 (P/F) ratio from 591 to 338 despite attempts to perform recruitment maneuvers and treatment with intravenous antibiotics and diuretics. Bronchoscopy appeared normal with only scant secretions and no evidence of aspiration. Gram stain of bronchoalveolar lavage revealed few gram negative rods and gram-positive cocci in pairs.

Donor met extended criteria donor based on the presence of two of the criteria (indicated by *) as below, with a lung donor score of 5:

Age> 55 years

Smoking history more than 20 pack-years

Chest trauma

Partial pressure of oxygen/fraction of inspired oxygen ratio < 300

Abnormal radiographic findings*

Abnormal bronchoscopy findings

Positive sputum microbiology*

Ischemic time > 6 hours

The lungs were accepted with anticipated use EVLP due to presence of atelectasis and declining P/F ratio. At the time of procurement, the PaO2/FiO2 ratio was 355. The lungs were placed on cold storage for transport to an independent, third party EVLP center. The lungs were inspected on arrival and placed on the EVLP circuit after 5 hours and 35 minutes of cold ischemic time (CIT #1). The lungs were monitored while perfused and ventilated on a normothermic circuit, utilizing the Toronto protocol and acellular perfusate.

Figure legend: (A-D): Red ovals show the areas of atelectasis, most marked in the right lower lobe at time of placement on EVLP circuit.

(E): After one hour of EVLP treatment, radiography shows persistent infiltrate suggestive that the right lower lobe remained atelectatic.

(F): After three hours of EVLP, follow up radiography shows significant improvement in the right lower lobe (RLL) opacity.

(G): Chest x-ray immediately post-bilateral lung transplant using donor lungs after EVLP procedure.

Final EVLP perfusate analysis showed a P/F ratio of 585. A subsequent deflation test (seen in video) shows effective recruitment of all lobes with slight, but acceptable delay in deflation only in the RLL. The lungs were accepted for lung transplant. The total duration of EVLP was 3 hours, 41 minutes. Lungs were successfully transplanted into the recipient with total ischemic time (CIT#1 + EVLP duration + CIT#2) of 11 hours and 53 minutes for the left lung and 12 hours and 58 minutes for the right lung. Immediately after transplant, the P/F ratio was 372 despite a hazy infiltrate noted on chest radiograph in the right lower lobe (G) which improved over the subsequent three days. Recipient was discharged home on post-transplant day 28.

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