**SUMMARY STATEMENT**

**PROGRAM CONTACT:** [redacted]

**( Privileged Communication )**

***Release Date:*** **04/10/2021**
***Revised Date:***

***Application Number:*** **1 K08 AI155816-01A1**
***Formerly:*** **1K08AI155816-01**

**Principal Investigator**

**AL-ADRA, DAVID**

**Applicant Organization: UNIVERSITY OF WISCONSIN-MADISON**

***Review Group:*** **AITC**
**Allergy, Immunology, and Transplantation Research Committee**
**Allergy, Immunology, and Transplantation Research Committee (AITC) February Council**

***Meeting Date:*** **02/25/2021**
***Council:*** **MAY 2021**
***Requested Start:*** **09/01/2021**

***RFA/PA:*** **PA20-203**
***PCC:*** **I5A**

***Project Title:*** **Targeting Donor Regulatory Dendritic Cells During Normothermic Ex Vivo Liver Perfusion to Overcome Rejection after Liver Transplant**

***SRG Action:*** **Impact Score:** [redacted]

***Next Steps:*** **Visit https://grants.nih.gov/grants/next_steps.htm**

**Human Subjects:** **10-No human subjects involved**

**Animal Subjects:** **30-Vertebrate animals involved - no SRG concerns noted**

| Project Year | Direct Costs Requested | Estimated Total Cost |
|---|---|---|
| 1 | [redacted] | [redacted] |
| 2 | | |
| 3 | | |
| 4 | | |
| 5 | | |
| TOTAL | [redacted] | [redacted] |

**ADMINISTRATIVE BUDGET NOTE: The budget shown is the requested budget and has not been adjusted to reflect any recommendations made by reviewers. If an award is planned, the costs will be calculated by Institute grants management staff based on the recommendations outlined below in the COMMITTEE BUDGET RECOMMENDATIONS section.**

**1K08AI155816-01A1 Al-Adra, D**

**RESUME AND SUMMARY OF DISCUSSION:** This exceptional resubmission Mentored Clinical Scientist Research Career Development Award application entitled “Targeting Donor Regulatory Dendritic Cells During Normothermic Ex Vivo Liver Perfusion to Overcome Rejection after Liver Transplant” was submitted in response to PA20-203: Mentored Clinical Scientist Research Career Development Award (Parent K08 Independent Clinical Trial Not Allowed) by the University of Wisconsin-Madison, Madison, Wisconsin with David Al-Adra, MD, PhD as Program Director/Principal Investigator (PD/PI).

Dr. Al-Adra is an outstanding candidate for the K08 award. He is an abdominal transplant surgeon and an expert in ex vivo normothermic liver perfusion, using a rat model, and he has excellent first author publications on this model. His clinical and research interests are well-aligned. His career development plan was criticized in the previous submission, and has now been revised and improved in this submission. Dr. Al-Adra will take courses in transplantation immunology, and he has found collaborators to assist with the rat liver transplantations. Dr. Al-Adra has an outstanding group of mentors who will advise him on the proposed research and his career development. The University of Wisconsin-Madison provides an excellent research environment and strong institutional support for Dr. Al-Adra. The research plan is improved from the previous submission and will address an important issue, liver transplant rejection, using an ex vivo normothermic perfusion model and a cytokine cocktail with TGF-beta and IL-10 during perfusion, which is hypothesized to expand regulatory dendritic cells (DCregs) and prevent transplant rejection. He will use an OX-62 antibody to deplete DCregs, although there is a lack of preliminary data with this antibody, raising concerns that the antibody may not be specific for DCregs. Overall, Dr. Al-Adra is an outstanding candidate who has addressed the previous concerns with his career development and research plan, and the proposed studies using a normothermic perfusion model could have a high impact and translational significance, aligning well with Dr. Al-Adra’s clinical interests and forming a strong foundation for Dr. Al-Adra’s independent research career.

Based upon the evaluation of scientific and technical merit, this application received an Impact/Priority score of [redacted]

**DESCRIPTION (provided by applicant):** ABSTRACT This proposal presents a five-year research career development program focused on targeting donor liver-resident cells with regulatory properties to decrease rejection after transplantation. I am an Assistant Professor of Surgery at the University of Wisconsin-Madison, with previous research and clinical experience in transplant immunology and transplant surgery involving normothermic ex vivo machine perfusion (NEVLP), whereby an organ is housed under physiologic conditions. The present project will advance the field of transplant immunology by using NEVLP technology to modify the immune cells within the liver prior to transplantation. I have assembled an outstanding mentorship team of investigators with expertise in transplant immunology, dendritic cell biology, and extracellular vesicle biology. The proposed training will guide and enhance my development in core competencies, including transplant immunology, communication, biostatistics, and ethical research design that will enable me to transition to research independence as a surgeon-scientist dedicated to reducing organ rejection in the field of transplant surgery. Liver transplantation is the only treatment option for patients with end-stage liver disease;

however, rejection of the transplant can decrease liver and patient survival. In addition, patients still require lifelong use of anti-rejection medications that suppress the immune system. Modification of the donor liver, and the immune cells within it, has the potential to promote acceptance of the liver and minimize the need for anti-rejection drugs. Advances in an innovative technique called normothermic ex vivo liver perfusion (NEVLP) offer a unique opportunity to benefit significantly the 25% of liver transplant recipients that develop acute rejection, as well as many more transplant recipients who would benefit from using fewer anti-rejection drugs. Recent studies have demonstrated the importance of regulatory dendritic cells (DCregs) for prolonging transplant survival. My central hypothesis is that expansion of the number of liver-resident DCregs during NEVLP will promote a regulatory environment for the organ after transplant. Using a rat model of NEVLP and liver transplantation that my research group has optimized, I expect NEVLP to expand DCregs potently, leading to an increase in immune checkpoint molecule expression and production of anti-inflammatory extracellular vesicles and cytokines that can reduce immune-mediated rejection. This innovative approach of expanding graft-resident DCregs to decrease rejection could be used in deceased donor liver transplantation as well as translated to other types of solid organ transplants. To achieve these objectives, I propose the following scientific aims:1) Determine the dominant regulatory function of liver-resident DCregs after NEVLP, and 2) Measure the impact of expanded liver-resident DCregs generated by combination cytokine therapy during NEVLP on liver graft rejection in vitro and in vivo.

**PUBLIC HEALTH RELEVANCE:** PUBLIC HEALTH RELEVANCE STATEMENT Transplant recipients must take lifelong anti-rejection medications that cause side effects and decrease their ability to defend against infections and cancers. We will use a novel method of liver storage in a rat model of liver transplantation to examine the impact of interventions aimed at increasing regulatory cells within the liver, thereby making the entire organ less likely to cause an immune reaction in the recipient. This exploration of strategies to decrease patient reliance on harmful drugs holds tremendous promise not only for liver transplantation, but all solid organ transplants.

**CRITIQUE:** The comments in the CRITIQUE section were prepared by the reviewers assigned to this application and are provided without significant modification or editing by staff. They are included to indicate the range of comments made during the discussion, and may not reflect the final outcome. The RESUME AND SUMMARY OF DISCUSSION section summarizes the final opinion of the committee after the discussion and is the basis for the assigned Overall Impact/Priority score.

**CRITIQUE 1**

| | |
|---|---|
| Candidate: | [redacted] |
| Career Development Plan/Career Goals /Plan to Provide Mentoring: | [redacted] |
| Research Plan: | [redacted] |
| Mentor(s), Co-Mentor(s), Consultant(s), Collaborator(s): | [redacted] |
| Environment Commitment to the Candidate: | [redacted] |

**Overall Impact:** This is a resubmission of a career development grant application that proposes to investigate novel methods to reduce rejection responses after liver transplantation. The initial submission had many strengths including an outstanding candidate with strong training in clinical transplantation and basic science, an outstanding team of mentors, an exceptional and highly supportive environment for the development of surgeon scientists and an innovative clinically relevant research proposal. Some weaknesses were identified, in particular with regard to the candidate's career development plan and the robustness of the preliminary data that supported his research proposal. In addition, it was felt that the application could benefit from more mechanistic studies. The applicant has been highly responsive to the previous critiques. Specifically, he presents a more refined career development plan with courses that fill his gap in knowledge, he presents more robust preliminary data and he has expanded the scope of his experiments to incorporate more mechanistic approaches. Some weaknesses remain in the research plan. For example, some of the proposed approaches do not specifically target DCregs (e.g. anti-PD-L1 treatment) and some approaches do not seem to have been validated (e.g. OX-62 treatment to deplete DCregs). Nevertheless, the strengths far outweigh the weaknesses and the level of enthusiasm for this application is high.

**1. Candidate:**

**Strengths**

- Very strong candidate with excellent training in clinical transplantation and basic science.
- Candidate has a PhD in Experimental Surgery.
- Candidate has several publications in recognized transplant journals.

**Weaknesses**

- None were noted.

**2. Career Development Plan/Career Goals and Objectives:**

**Strengths**

- In response to previous critiques, the candidate has revised and clarified his training plan.
- In response to previous critiques, the candidate includes a letter indicating support from other laboratory with establishing microsurgical models.
- Candidate's clinical and research areas are highly complementary.

**Weaknesses**

- None were noted.

**3. Research Plan:**

**Strengths**

- Clinically relevant and significant area of investigation
- Expanding regulatory cell populations during normothermic ex vivo perfusion is considered to be conceptually innovative.
- Physiological model of liver transplantation in rodent model with defined strain combinations that either reject or spontaneously accept livers.

- In response to previous critiques, the applicant has expanded the preliminary data that support the studies. Specifically, he has increased the n's for some figures and also shows flow cytometry plots outlining gating scheme.

**Weaknesses**

- Some of the approaches still lack specificity for DCregs (e.g. administration of anti-PD-L1 antibody in Aim 1). Applicant acknowledges some of these limitations.
- Applicant proposes to use OX-62 antibodies to deplete DCregs. Ideally, the ability to deplete this cell population with this specific antibody needs to be shown to increase confidence in this approach.

**4. Mentor(s), Co-Mentor(s), Consultant(s), Collaborator(s):**

**Strengths**

- Outstanding mentoring team consisting of recognized experts whose areas of expertise are highly complementary.

**Weaknesses**

- None were noted.

**5. Environment & Institutional Commitment to the Candidate:**

**Strengths**

- Exceptional environment for the development of surgeon scientists, especially in transplantation. Several of the applicant's colleagues in his division are surgeon scientists.
- Very strong institutional support
- Letters of support indicate that the candidate will have 75% protected time irrespective of whether his K08 application is successful.

**Weaknesses**

- None were noted.

**CRITIQUE 2**

| | |
|---|---|
| Candidate: | [redacted] |
| Career Development Plan/Career Goals /Plan to Provide Mentoring: | [redacted] |
| Research Plan: | [redacted] |
| Mentor(s), Co-Mentor(s), Consultant(s), Collaborator(s): | [redacted] |
| Environment Commitment to the Candidate: | [redacted] |

**Overall Impact:** This K08 grant application is an A1 submission by Dr. David Al-Adra to develop his basic research career in immunobiology of liver transplantation and the mechanism of acute liver transplant rejection. Dr. Al-Adra's long time goal is to ultimately translate research findings in this and future projects to develop therapy to prevent acute rejection in orthotopic liver transplantation (OLT) and potentially also prevent in other types of solid organ transplantation.

This grant application is highly likely to lead to successful development of Dr. David Al-Adra into an independent surgeon scientist with a basic and translational science research program in the prevention of acute rejection in OLT. The main strengths of this grant application are: (1) an exceptionally well-prepared candidate with a PhD in Experimental Surgery and high level of productivity including 34 peer-reviewed publications of which 11 were first authored; (2) a comprehensive career development plan that combines essential didactic training necessary to fill the gaps identified in Dr. Al-Adra's training and lead to the acquisition of laboratory skills and training in ethics, management and leadership in clinical research that are necessary for the current project; (3) an elegant and innovative research hypothesis with significant changes to address A0 critiques in the research plan; (4) substantial institutional commitment with 75% protected time and fully equipped and staffed laboratory and (5) a multidisciplinary mentoring team with complementary expertise. There are no major weaknesses in this grant application. The enthusiasm for this application is very high.

**1. Candidate:**

**Strengths**

- Dr. Al-Adra is well-prepared for this K08 grant application with more than 15 years of prior research training and a PhD in Experimental Surgery.
- Dr. Al-Adra has 34 publications including 12 publications in transplant immunobiology and 11 of all publications were first-authored publications.
- Excellent cutting-edge research training from the University of Alberta and the University of Toronto including pioneering training experience in the application of NEVLP in North America.

**Weaknesses**

- None noted.

**2. Career Development Plan/Career Goals and Objectives:**

**Strengths**

- Didactic and laboratory skills training are tailored to fill the gaps in prior training.
- Biostatistics, ethics, research lab management and leadership are appropriately featured in the career development plan.
- Monitoring and evaluation components are comprehensive and feasible.

**Weaknesses**

- None were noted.

**3. Research Plan:**

**Strengths**

- The experiments to achieve the two specific aims were redesigned to explore how DCregs reduce rejection and mechanisms of DCregs expansion with NEVLP and anti-inflammatory cytokines.
- Potential problems and alternative approaches have been adequately addressed.
- The research plan is appropriate for the level of career development of Dr. Al-Adra.
- The plan to perform NEVLP and the use of various rat models of orthotopic liver transplantation are rigorous and well-written.

**Weaknesses**

- None were noted.

**4. Mentor(s), Co-Mentor(s), Consultant(s), Collaborator(s):**

**Strengths**

- Dr. Christian Capitini has the research and mentoring experience to serve as a mentor for Dr. Al-Adra
- Dr. Willian Burlingham's experience and expertise will complement that of Dr. Capitini. Dr. Burlingham is an internationally renowned senior research scientist in transplant immunology. He has extensive mentoring experience with physician scientists.
- Dr. Paul Sondel and Dr. Josh Mezrich are highly qualified to serve as co-mentors respectively.

**Weaknesses**

- None were noted.

**5. Environment & Institutional Commitment to the Candidate:**

**Strengths**

- The research environment at the University of Wisconsin, Madison is superlatively rich in research resources and it is one of the foremost transplant centers in the world. It offers the best intellectual milieu possible for the work proposed in this application.
- The letter of commitment from Dr. Rebecca Minter, Chair, Department of Surgery is very strong. The start-up package is phenomenally high and protected time given to Dr. Al-Adra is generous.

**Weaknesses**

- None were noted.

**CRITIQUE 3**

| | |
|---|---|
| Candidate: | [redacted] |
| Career Development Plan/Career Goals /Plan to Provide Mentoring: | [redacted] |
| Research Plan: | [redacted] |

Mentor(s), Co-Mentor(s), Consultant(s), Collaborator(s): [redacted]

Environment Commitment to the Candidate: [redacted]

**Overall Impact:** This revised application is quite responsive to the prior reviewer critiques of the project. The career development plan and research plan are more thoroughly detailed. In particular, the research plan is significantly expanded and is designed to address numerous important questions regarding DCregs and their potential expansion during NEVLP to promote tolerance. Characterization of the phenotype of DCregs generated in various experimental conditions could provide additional insight into their function. Overall this is an excellent project that is well suited for Dr. Al-Adra's career trajectory as a transplant surgeon-scientist.

**1. Candidate:**

**Strengths**

- Dr. Al-Adra is a surgeon-scientist with an MD-PhD. He has focused on transplant immunology throughout his career.
- Solid publication record

**Weaknesses**

- None

**2. Career Development Plan/Career Goals and Objectives:**

**Strengths**

- Reasonable career development plan

**Weaknesses**

- None.

**3. Research Plan:**

**Strengths**

- Both aims are now designed to investigate two key features in the NEVLP system: (1) effect of expanding or depleting DCregs (or DCreg-derived extracellular vesicles) upon liver tolerance and rejection; and (2) modulating the cytokine microenvironment during NEVLP to expand DCregs.
- Experiments are more thoroughly described and anticipated outcomes of each experiment are clear.

**Weaknesses**

- In addition to examining the effects of DCreg expansion or inhibition, characterization of the phenotype of the DCregs that arise under the various experimental conditions could be useful, to identify those that may be most beneficial for clinical transplantation.

**4. Mentor(s), Co-Mentor(s), Consultant(s), Collaborator(s):**

**Strengths**

- Very strong mentorship team

**Weaknesses**

- None

**5. Environment & Institutional Commitment to the Candidate:**

**Strengths**

- Excellent institutional commitment

**Weaknesses**

- None

**Resubmissions:**
Comments or Concerns: ACCEPTABLE

**THE FOLLOWING RESUME SECTIONS WERE PREPARED BY THE SCIENTIFIC REVIEW OFFICER TO SUMMARIZE THE OUTCOME OF DISCUSSIONS OF THE REVIEW COMMITTEE ON THE FOLLOWING ISSUES:**

**PROTECTION OF HUMAN SUBJECTS: NOT APPLICABLE (CODE 10)**

**VERTEBRATE ANIMAL: ACCEPTABLE (CODE 30)**
The protection of Vertebrate Animal welfare is adequately described.

**BIOHAZARD COMMENT: ACCEPTABLE**
The plan to prevent risks during handling of biohazard materials or samples is adequate.
BSL-2: Biosafety cabinets described.

**TRAINING IN THE RESPONSIBLE CONDUCT OF RESEARCH: ACCEPTABLE**
All 5 elements are adequately addressed in the application.
**Format:** Acceptable
**Subject Matter:** Acceptable
**Faculty Participation:** Acceptable
**Duration:** Acceptable
**Frequency:** Acceptable

**SELECT AGENT: NOT APPLICABLE**

**RESOURCE SHARING PLANS**

**DATA SHARING PLAN: NOT APPLICABLE**

**MODEL ORGANISMS SHARING PLAN: NOT APPLICABLE**

**GENOMIC DATA SHARING PLAN: NOT APPLICABLE**

**FOREIGN INSTITUTION: N/A**

**AUTHENTICATION OF KEY BIOLOGICAL AND/OR CHEMICAL RESOURCES: ACCEPTABLE**
Adequate plans for authentication of key resources are outlined in the application.

**BUDGETARY OVERLAP: NOT APPLICABLE**

**COMMITTEE BUDGET RECOMMENDATIONS:**
The budget is recommended as requested in all years.

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Footnotes for 1 K08 AI155816-01A1; PI Name: Al-Adra, David

NIH has modified its policy regarding the receipt of resubmissions (amended applications).See Guide Notice NOT-OD-18-197 at https://grants.nih.gov/grants/guide/notice-files/NOT-OD-18-197.html. The impact/priority score is calculated after discussion of an application by averaging the overall scores (1-9) given by all voting reviewers on the committee and multiplying by 10. The criterion scores are submitted prior to the meeting by the individual reviewers assigned to an application, and are not discussed specifically at the review meeting or calculated into the overall impact score. Some applications also receive a percentile ranking. For details on the review process, see http://grants.nih.gov/grants/peer_review_process.htm#scoring.