AWARD NUMBER: W81XWH-16-1-0680

TITLE: MIF-Based Therapies in Cigarette Smoke-Related COPD and Pneumonia

PRINCIPAL INVESTIGATOR: Patty Lee, MD

RECIPIENT: Yale University
New Haven,CT 06511

REPORT DATE: October 2017

TYPE OF REPORT: Annual

PREPARED FOR: U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1. REPORT DATE	2. REPORT TYPE	3. DATES COVERED
October 2017	Annual	15 Sep 2016 - 14 Sep 2017

4. TITLE AND SUBTITLE

MIF-Based Therapies in Cigarette Smoke-Related COPD and Pneumonia

5a. CONTRACT NUMBER

5b. GRANT NUMBER
W81XWH-16-1-0680

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)

Patty Lee, M.D.

E-Mail: patty.lee@yale.edu

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)

Yale University
Department of Pulmonary and Critical Care Medicine,
MIMED 721774
15 York Street , New Haven, CT 06510-3221

8. PERFORMING ORGANIZATION REPORT

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)

U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

10. SPONSOR/MONITOR'S ACRONYM(S)

11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION / AVAILABILITY STATEMENT

Approved for Public Release; Distribution Unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT

Chronic obstructive pulmonary disease (COPD) is the 3rd leading cause of death worldwide and is especially common in military members and Veterans. A major cause of mortality in people with COPD is bacterial pneumonia caused by *Streptococcus pneumoniae* (*S. pneumoniae*). We identified an innate immune protein, Macrophage migration inhibitory factor (MIF) and it receptor, CD74, as endogenous, protective molecules that determine susceptibility to COPD and immunity against *S. pneumoniae*. Augmenting MIF levels in susceptible individuals may be effective therapy against COPD as well as *S. pneumoniae* infection. We have already developed orally active MIF agonists (MIF20) with excellent safety and efficacy profiles that are ready to be tested in COPD and bacterial pneumonia models. **Our overall objective of this proposal is test MIF augmentation against CSE-related COPD and its subsequent complication, *S. pneumoniae*.** We propose to complete the following two **Aims**: **1)** Test the therapeutic efficacy of MIF augmentation in CSE-related COPD; **2)** Test the therapeutic efficacy of MIF augmentation in CSE-related *S. pneumoniae* infection. These studies will provide pre-clinical testing of MIF agonist-based therapy in COPD and bacterial pneumonia.

15. SUBJECT TERMS

Lungs, COPD, emphysema, cigarette smoke, pneumonia, macrophage inhibitory factor, inflammation, immunity, therapy

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	c. THIS PAGE			USAMRMC
Unclassified	Unclassified	Unclassified	Unclassified	17	19b. TELEPHONE NUMBER *(include area code)*

TABLE OF CONTENTS

	Page
1. Introduction	4
2. Keywords	4
3. Accomplishments	4-9
4. Impact	9-10
5. Changes/Problems	10-11
6. Products	11-14
7. Participants & Other Collaborating Organizations	14-17
8. Special Reporting Requirements	N/A
9. Appendices	N/A

1. **INTRODUCTION:** Narrative that briefly (one paragraph) describes the subject, purpose and scope of the research.

COPD is a progressive, destructive process of airflow obstruction, leading to respiratory failure. A major cause of mortality in people with COPD is bacterial pneumonia, such as those caused by *Streptococcus pneumonia* (*S. pneumoniae*). The main purpose of this research project is to perform pre-clinical therapeutic testing of MIF augmentation strategies, leveraging our recently-developed small molecules, in a mouse model of human COPD and bacterial pneumonia.

2. **KEYWORDS:** Provide a brief list of keywords (limit to 20 words).

Lungs, COPD, emphysema, cigarette smoke, pneumonia, macrophage inhibitory factor, inflammation, immunity, therapy

3. **ACCOMPLISHMENTS:** The PI is reminded that the recipient organization is required to obtain prior written approval from the awarding agency grants official whenever there are significant changes in the project or its direction.

What were the major goals of the project?
List the major goals of the project as stated in the approved SOW. If the application listed milestones/target dates for important activities or phases of the project, identify these dates and show actual completion dates or the percentage of completion.

Major Goals/Milestones:

1. HRPO & ACURO Approval: 100% completed
2. Yale IRB/IACUC Approval: 100% completed
3. Dose selection using 3 doses and 3 routes of delivery (oral, systemic and intra-tracheal) and check for overt toxicity after 1 month. 100% completed
4. 1 dose selected and no toxicity detected: 100% completed
5. Increased activation and MIF-related proteins after MIF20 administration: 100% completed
6. Decreased injury and inflammation after MIF20 administration: 50% completed
7. Therapeutic effect after short-term smoke exposure in at least 2 of the 3 routes: 20% completed

What was accomplished under these goals?
For this reporting period describe: 1) major activities; 2) specific objectives; 3) significant results or key outcomes, including major findings, developments, or conclusions (both positive and negative); and/or 4) other achievements. Include a discussion of stated goals not met. Description shall include pertinent data and graphs in sufficient detail to explain any significant results achieved. A succinct description of the methodology used shall be provided. As the project progresses to completion, the emphasis in reporting in this section should shift from reporting activities to reporting accomplishments.

These goals allowed the testing and selection of the appropriate dose and route of administration of MIF20, a MIF agonist, using mice. Major activities included testing of the solubility of MIF20 in various diluents to achieve safe in vivo delivery. The doses, routes of administration and results are shown in Figures 1 – 7. These results allowed us to determine the impact of 3 routes of delivery on lung histology and protein expression. In addition, we could determine whether a gender effect was noted, using 50% male and 50% female mice for all experiments. Initial challenges that had to be overcome prior to obtaining these results were the insolubility of MIF20, which required multiple different diluents at varying concentrations. We eventually selected a dose of Tocrisolve that achieved solubility but found initial lung inflammation in the mice but eventually identified a dose of MIF20 and concentration of MIF20 that appeared to be tolerated by oral gavage. We had also initially attempted to nebulize MIF20 but due to the adherence of the MIF20 in Tocrisolve to the nebulization apparatus, we tested the lung-targeted route using direct intra-tracheal instillation.

1. **Intra-peritoneal (IP) injection**
   - 40 mg/kg/7.5 mL
   - Vehicle: 21.1% Tocrisolve 100 in saline
2. **Intra-tracheal (IT) injection**
   - 80 mg/kg/2 mL
   - Vehicle: 100% Tocrisolve 100
3. **Intra-gastric (IG) injection**
   - 100 mg/kg/10 mL
   - Vehicle: 1% CMC in water

Figure 1. Experimental details. Animals: C57BL/6 mice (female and male, 6 weeks old) were supplied by Jackson Laboratory (Bar Harbor, ME, USA). The animals were housed in cages located in temperature- and humidity-controlled rooms with a 12-h light-dark cycle and received water and food ad libitum. **MIF20 administration and experimental design:** Mice were intra-peritoneally injected vehicle (21% Tocrisolve 100; Tocris Bioscience, Bristol, UK) or 40 mg/kg MIF20 once a day for 4 weeks. Mice were intra-tracheally injected vehicle (Tocrisolve 100) or 80 mg/kg MIF20 once a day for 4 weeks. Mice were orally injected vehicle (1% carboxymethyl cellulose) or 100 mg/kg MIF20 once a day for 4 weeks. Body weight was recorded for up to 4 weeks (each group n=4). Under anesthesia with urethane, lung tissues were collected. **Total protein extraction:** The lung tissues were homogenized in M-PER (Thermo Fisher Scientific, Waltham, MA, USA) for total protein samples according to the manufacturer's instruction. The protein concentration was determined using BCA Protein Assay kit (Thermo Fisher Scientific). **Western blot analysis:** Protein samples (12-16 ug) were loaded on 4-15% gradient TGX gel (Bio-Rad Laboratories, Hercules CA, USA), separated by sodium dodecyl sulphate/polyacrylamide gel electrophoresis and transferred to polyvinylidene fluoride membranes (Millipore, Billerica, MA, USA) using the Semi-Dry Trans-Blot Cell (Bio-Rad Laboratories). After the transfer, the membranes were blocked for 1-h with 5% (w/v) skim milk or bovine serum albumin powder in Tris-buffered saline with 0.1% Tween-20 at room temperature. Blots were incubated with primary antibodies overnight at 4°C. Primary antibodies against p-AKT, AKT, p-ERK, ERK (Cell Signaling Technology, Danvers, MA, USA), p-p38 and p38 (Santa Cruz Biotechnology, Dallas, TX, USA) were used. On the following day, the blots were incubated with appropriate secondary antibodies for 1-h at room temperature. Bands were detected using an ECL detection system (iNtRON Biotechnology) according to the manufacturer's instructions. The intensities of immunoreactive bands were evaluated using Total-Lab TL120 software (Nonlinear Dynamics, Newcastle, UK). Signals were standardized to that of β-actin (Sigma-Aldrich, St. Louis, MO, USA). **Statistical analysis:** All data were analyzed by the student t-test was used for post-hoc comparisons. Statistical differences between the groups were considered significant at $p<0.05$.

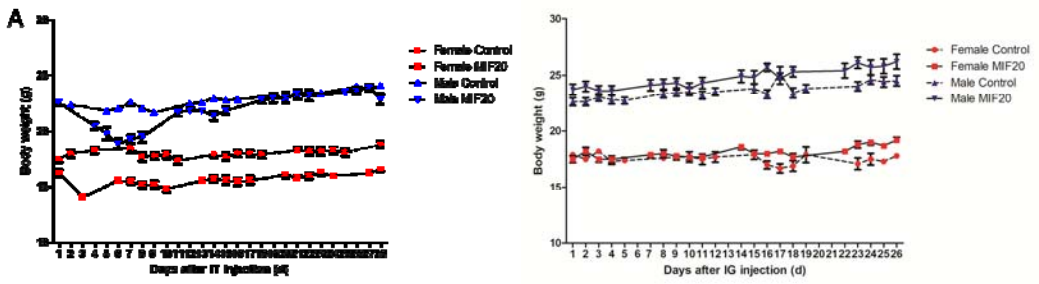

Figure 2. The effect of MIF20 on body weight. The mice were given intra-tracheal injection of vehicle (Tocrisolve 100; Tocris Bioscience) or MIF20 80 mg/kg for 4 weeks **(A)**. The mice were given oral injection of vehicle (1% carboxymethyl cellulose) or MIF20 100 mg/kg for 4 weeks **(B)**.

Interpretation & Plans: The logarithm of body weight is related to straight-line relationship with the logarithm of the LD_{50} (Toxicol Appl Pharmacol, 1968). Thus, body weight is considered as the primary and gold standard in toxicology research as a sign of general toxicity. In the present study, the mortality, change in body weight and gross observation were monitored for 28 days during MIF20 treatment. As suggested by the results, we did not detect any mortality, changes in the body weight and gross findings of toxicity in IT-injected male mice and IG-injected both female and male mice. The body weights detected in these mice corresponded well to the body weight ranges of age-matched normal mice (http://www.jax.org). Of note, IT treatment of MIF20 induced loss of body weight during the first 1 week, which may indicate the presence of side effects that require acclimation. However, after the first week, the male mice started to recover their body weight to normal range, while the female mice showed persistent, decreased body weight, which suggests there may be gender differences in the way MIF20 is metabolized or its physiologic effects. Based on these results, we will proceed with MIF20 administration but monitor body weight and gender-related differences in MIF20 cellular/molecular effects as we advance to the next round of testing.

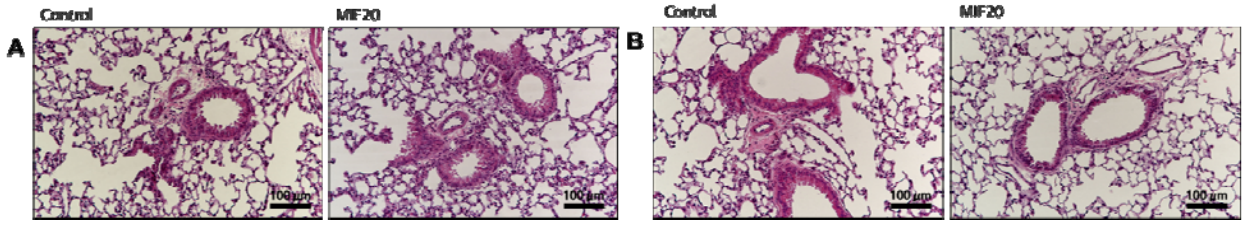

Figure 3. The effect of MIF20 on lung histology. The mice (**A:** female, **B:** male) were given intra-peritoneal injection of vehicle (21% Tocrisolve 100; Tocris Bioscience) or MIF20 40 mg/kg for 4 weeks. After clearing the pulmonary intravascular space with ice-cold PBS, whole lungs were dissected from mice and immediately fixed in 4% paraformaldehyde for overnight at 4°C. After paraffin embedding, histological analysis was performed on 5 um sections and stained with hematoxylin and eosin by the Yale Histopathology Services. Histological changes were evaluated in random and nonconsecutive fields.

Interpretation: Hematoxylin and eosin stain is one of the principal stains in histology. The histological structure of the lung parenchyma and air duct is normal in IP-injected mice group. These results assure us that MIF20 is not leading to overt airway or lung parenchymal cellular inflammation when administered systemically, via IP injection.

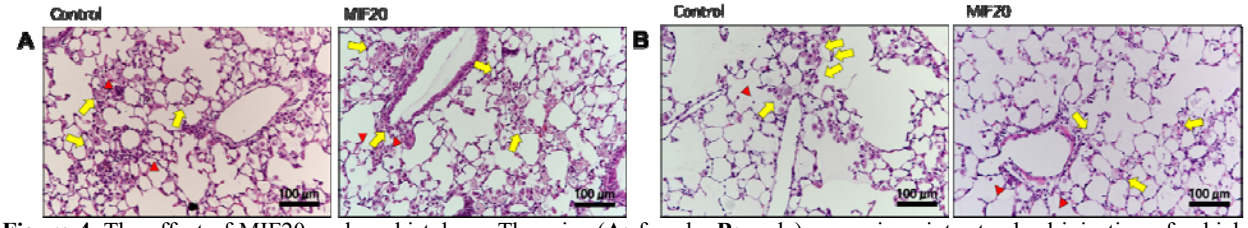

Figure 4. The effect of MIF20 on lung histology. The mice (**A:** female, **B:** male) were given intra-tracheal injection of vehicle (Tocrisolve 100; Tocris Bioscience) or MIF20 80 mg/kg for 4 weeks. After clearing the pulmonary intravascular space with ice-cold PBS, whole lungs were dissected from mice and immediately fixed in 4% paraformaldehyde for overnight at 4°C. After paraffin embedding, histological analysis was performed on 5 um sections and stained with hematoxylin and eosin by the Yale Histopathology Services. Histological changes were evaluated in random and nonconsecutive fields. Yellow and red arrows indicate macrophage and neutrophil, respectively.

Interpretation & Plans: IT treatment of vehicle and MIF20 induced severe inflammatory cell infiltration in lungs. Both neutrophils and macrophages were accumulated into airway. Of note, inflammatory cell infiltration was induced by not only MIF injection but also vehicle (Tocrisolve 100) injection. Moreover, some of macrophages engulfed the debris and/or particles. Tocrisolve 100 is composed of a 1:4 ratio of soya oil/water that is emulsified with the block co-polymer Pluronic F68. Dispersing agents (e.g., Pluronic F68) are sometimes added to the saline/vehicle to keep suspended particles from agglomerating or settling in IT injection. However, such agents can disrupt endogenous surfactant and can even elicit adverse effects of their own (Toxicol Appl Pharmacol, 1982; Am J Respir Crit Care Med, 1997). Although it was reported that intratracheal instillation of recombinant MIF increased the number of neutrophils in BAL fluids (Respir Res, 2009), we did not find differences in infiltrated neutrophil numbers between control and MIF20 groups. We concluded that IT injection is not an ideal administration route for MIF20 unless an alternative solvent is used.

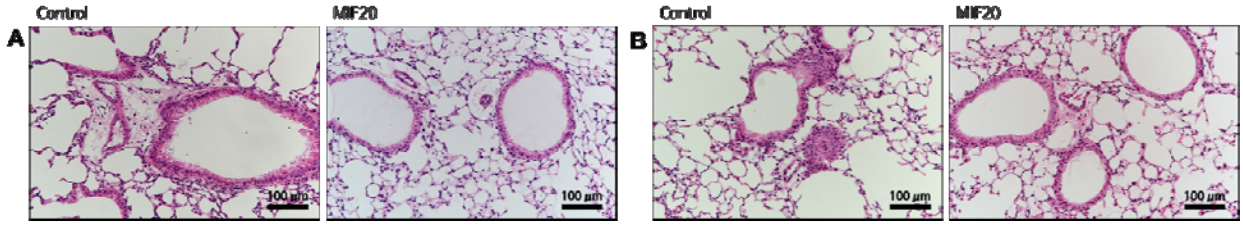

Figure 5. The effect of MIF20 on lung histology. The mice (**A:** female, **B:** male) were given oral injection of vehicle (1% carboxymethyl cellulose) or MIF20 100 mg/kg for 4 weeks. After clearing the pulmonary intravascular space with ice-cold PBS, whole lungs were dissected from mice and immediately fixed in 4% paraformaldehyde for overnight at 4°C. After paraffin embedding, histological analysis was performed on 5 um sections and stained with hematoxylin and eosin by the Yale Histopathology Services. Histological changes were evaluated in random and nonconsecutive fields.

Interpretation & Plans: The histological structure of the lung parenchymal and air duct is normal in IG-injected mice group. There is no inflammatory cell infiltration around the airway. We concluded that IG may be preferred over direct lung instillation (IT).

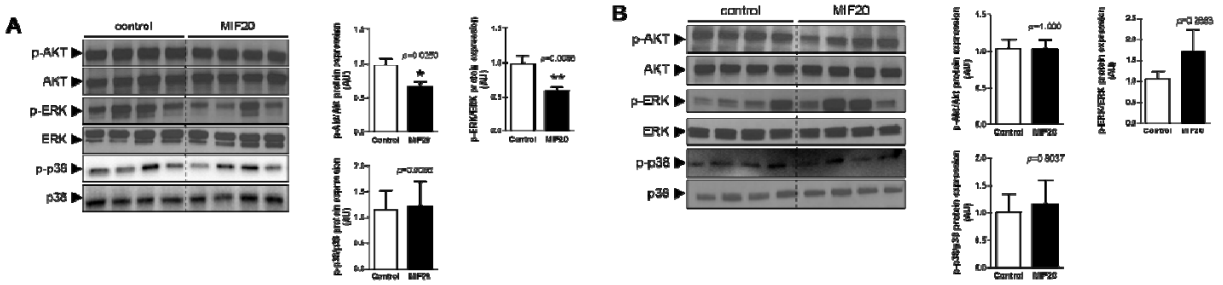

Figure 6. The effect of MIF20 on the expression of MIF signaling-related proteins. The mice (**A:** female, **B:** male) were given intra-peritoneal injection of vehicle (21% Tocrisolve 100; Tocris Bioscience) or MIF20 80 mg/kg for 4 weeks. The lung tissues were collected at 4-weeks and then p-AKT, p-ERK and p-p38 protein expression were determined. The results are presented as mean ± SEM (n=4 per group). *p<0.05, **p<0.01 versus control group.

Interpretation & Plans: MIF is a structurally unique innate cytokine that is present in preformed cytoplasmic pools and is rapidly released in response to microbial components or oxidants, where its main action is to sustain cell survival (Current Immunology Reviews, 2006). It has been shown that CD74 was the cell surface receptor for MIF, and that MIF promotes sustained ERK/MAPK activation through binding CD74 receptor (Cell Signal, 2004; Cell Signal, 2006). In addition, MIF can activate the PI3K/AKT pathway, as well as promote endothelial cell proliferation and differentiation (Oncogene, 2007; Circ Res, 2003). To determine the effect of MIF20 on its known downstream signaling proteins, p-AKT, p-ERK and p-p38, we proceeded with Western blots. IP injection of MIF20 for 4 weeks significantly decreased p-AKT and p-ERK protein expressions (68.2% and 59.5% of those in the control group, respectively) in female mice. There was no significant difference between the groups in p-p38 protein expression. Zhang et al. reported that MIF overexpression statistically decreased the protein levels of p-ERK1/2, p-PI3K and p-AKT, while MIF exerted no significant effect on the protein levels of EKR1/2, PI3K and AKT (Neuroscience, 2017). Thus, we will reduce the dose of MIF20 in future studies. We also determined there were no significant differences between the groups in p-AKT, p-EKR and p-p38 protein expression in male mice. Some reports described a relationship between sex hormones and MIF levels. For example, MIF levels in colon increased significantly from the follicular to the luteal phase of the cycles, which means estrogen decreased MIF production, while progesterone increased its production (Gastroenterology, 2007). In addition, MIF concentrations were significantly decreased in ovariectomized female mice. Therefore, we concluded that we will attempt to control for sex hormone differences by factoring in gender in our analyses and by using same-aged female mice. In female mice, we will also inject MIF20 for 5 consecutive days to cover the full estrous cycle (average estrous cycle for mouse is 4 to 5 days).

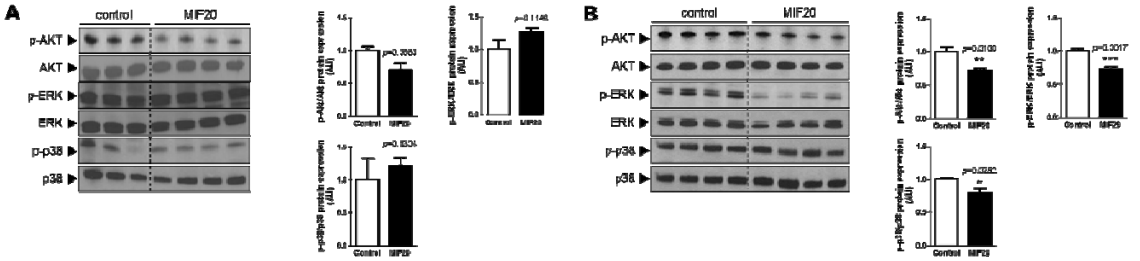

Figure 7. The effect of MIF20 on the expression of MIF signaling-related proteins. The mice were given oral injection of vehicle (1% carboxymethyl cellulose) or MIF20 100 mg/kg for 4 weeks. The lung tissues were collected at 4-weeks and then p-AKT, *p*-ERK and p-p38 protein expression were determined. The results are presented as mean ± SEM (n=4 per group). **p*<0.05, ***p*<0.01, ****p*<0.001 versus control group.

Interpretation & Plans: IG injection of MIF20 for 4 weeks significantly decreased p-AKT, p-ERK and p-p38 protein expressions (72.6%, 73.5% and 78.8% of those in the control group, respectively) in male mice. There were no significant differences of these protein expression between the groups in female mice. In the next set of experiments, MIF20 dose will be reduced and p-AKT, p-ERK and p-p38 protein levels in lung tissues will be measured to select optimal dose of MIF20.

What opportunities for training and professional development has the project provided?

If the project was not intended to provide training and professional development opportunities or there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe opportunities for training and professional development provided to anyone who worked on the project or anyone who was involved in the activities supported by the project. "Training" activities are those in which individuals with advanced professional skills and experience assist others in attaining greater proficiency. Training activities may include, for example, courses or one-on-one work with a mentor. "Professional development" activities result in increased knowledge or skill in one's area of expertise and may include workshops, conferences, seminars, study groups, and individual study. Include participation in conferences, workshops, and seminars not listed under major activities.

These studies resulted in new training opportunities by 1) promoting proficiency in medicinal chemistry and pharmacology, as necessitated by the manipulation of MIF20 in various diluents and concentrations, 2) proficiency in toxicity-testing, as necessitated by daily observations of mouse behavior, body weight measurements and lung histologic analyses, 3) proficiency in mouse handling and 4) proficiency in lung protein measurements.
Professional development activities include broadening of scientific knowledge in lung biology, initiating new interactions with pharmaceutical colleagues (who provide MIF20) and the presentation of the results at Pulmonary research lab meetings and conferences.

How were the results disseminated to communities of interest?

If there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe how the results were disseminated to communities of interest. Include any outreach activities that were undertaken to reach members of communities who are not usually aware of these project activities, for the purpose of enhancing public understanding and increasing interest in learning and careers in science, technology, and the humanities.

Nothing to Report

What do you plan to do during the next reporting period to accomplish the goals?
If this is the final report, state "Nothing to Report."

Describe briefly what you plan to do during the next reporting period to accomplish the goals and objectives.

The next set of goals and milestones to achieve are the following:

1. Complete the short-term cigarette smoke exposures.
2. Initiate the long-term cigarette smoke exposures.
3. Initiate MIF- genotype and MIF plasma studies.
4. Initiate bacterial pneumonia challenges.

4. **IMPACT:** Describe distinctive contributions, major accomplishments, innovations, successes, or any change in practice or behavior that has come about as a result of the project relative to:

What was the impact on the development of the principal discipline(s) of the project?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe how findings, results, techniques that were developed or extended, or other products from the project made an impact or are likely to make an impact on the base of knowledge, theory, and research in the principal disciplinary field(s) of the project. Summarize using language that an intelligent lay audience can understand (Scientific American style).

Our detailed testing of 3 delivery routes in mice, using different doses of MIF20, will expand our base of knowledge about lung-targeted drug-delivery options and deepen our understanding of how MIF-based strategies may or may not be used to treat cigarette smoke-related lung diseases.

What was the impact on other disciplines?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe how the findings, results, or techniques that were developed or improved, or other products from the project made an impact or are likely to make an impact on other disciplines.

Our studies will impact the field of lung-targeted pharmacology and drug-development and given that cigarette smoke negatively impacts most other organs, in addition to lung, such as brain, heart, eyes, pancreas, liver and gut, our studies on the potential therapeutic potential of MIF will have wide-ranging effects on many other disciplines. In addition, our studies will be one of the few of its kind to test MIF strategies small molecules in a model of bacterial pneumonia after smoke-exposure.

What was the impact on technology transfer?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe ways in which the project made an impact, or is likely to make an impact, on commercial technology or public use, including:

- *transfer of results to entities in government or industry;*
- *instances where the research has led to the initiation of a start-up company; or*
- *adoption of new practices.*

We are performing fundamental in vivo testing of an immune modulator in a complex, chronic lung process for which no specific therapies exist and therefore, these studies are critical to future drug-development for severe, chronic lung disease.

What was the impact on society beyond science and technology?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe how results from the project made an impact, or are likely to make an impact, beyond the bounds of science, engineering, and the academic world on areas such as:

- *improving public knowledge, attitudes, skills, and abilities;*
- *changing behavior, practices, decision making, policies (including regulatory policies), or social actions; or*
- *improving social, economic, civic, or environmental conditions.*

If these studies identify MIF as a therapy or as a diagnostic tool, the practice of medicine and delivery of treatment against cigarette smoke-related diseases as well as bacterial pneumonia would be significantly altered.

5. CHANGES/PROBLEMS: The PD/PI is reminded that the recipient organization is required to obtain prior written approval from the awarding agency grants official whenever there are significant changes in the project or its direction. If not previously reported in writing, provide the following additional information or state, "Nothing to Report," if applicable:

Nothing to Report

Describe any changes in approach during the reporting period and reasons for these changes. Remember that significant changes in objectives and scope require prior approval of the agency.

Actual or anticipated problems or delays and actions or plans to resolve them
Describe problems or delays encountered during the reporting period and actions or plans to resolve them.

Nothing to Report

Changes that had a significant impact on expenditures
Describe changes during the reporting period that may have had a significant impact on expenditures, for example, delays in hiring staff or favorable developments that enable meeting objectives at less cost than anticipated.

The original allocated salary for the operation of the smoking apparatus, Teague, had to be increased due to the level of training required. The original salary allocation was for the level of a postgraduate but the complexity and technical skills required is the level of a research associate.

Significant changes in use or care of human subjects, vertebrate animals, biohazards, and/or select agents

Describe significant deviations, unexpected outcomes, or changes in approved protocols for the use or care of human subjects, vertebrate animals, biohazards, and/or select agents during the reporting period. If required, were these changes approved by the applicable institution committee (or equivalent) and reported to the agency? Also specify the applicable Institutional Review Board/Institutional Animal Care and Use Committee approval dates.

Significant changes in use or care of human subjects

Nothing to Report

Significant changes in use or care of vertebrate animals

Nothing to Report

Significant changes in use of biohazards and/or select agents

Nothing to Report

6. **PRODUCTS:** List any products resulting from the project during the reporting period. If there is nothing to report under a particular item, state "Nothing to Report."

- **Publications, conference papers, and presentations**

  Report only the major publication(s) resulting from the work under this award.

  Journal publications. *List peer-reviewed articles or papers appearing in scientific, technical, or professional journals. Identify for each publication: Author(s); title; journal; volume: year; page numbers; status of publication (published; accepted, awaiting publication; submitted, under review; other); acknowledgement of federal support (yes/no).*

Nothing to Report

  Books or other non-periodical, one-time publications. *Report any book, monograph, dissertation, abstract, or the like published as or in a separate publication, rather than a*

periodical or series. Include any significant publication in the proceedings of a one-time conference or in the report of a one-time study, commission, or the like. Identify for each one-time publication: author(s); title; editor; title of collection, if applicable; bibliographic information; year; type of publication (e.g., book, thesis or dissertation); status of publication (published; accepted, awaiting publication; submitted, under review; other); acknowledgement of federal support (yes/no).

Nothing to Report

Other publications, conference papers and presentations. *Identify any other publications, conference papers and/or presentations not reported above. Specify the status of the publication as noted above. List presentations made during the last year (international, national, local societies, military meetings, etc.). Use an asterisk (*) if presentation produced a manuscript.*

Nothing to Report

- **Website(s) or other Internet site(s)**
  List the URL for any Internet site(s) that disseminates the results of the research activities. A short description of each site should be provided. It is not necessary to include the publications already specified above in this section.

Nothing to Report

- **Technologies or techniques**
  Identify technologies or techniques that resulted from the research activities. Describe the technologies or techniques were shared.

Nothing to Report

- **Inventions, patent applications, and/or licenses**
  Identify inventions, patent applications with date, and/or licenses that have resulted from the research. Submission of this information as part of an interim research performance progress report is not a substitute for any other invention reporting required under the terms and conditions of an award.

Nothing to Report

- **Other Products**
  Identify any other reportable outcomes that were developed under this project. Reportable outcomes are defined as a research result that is or relates to a product, scientific advance, or research tool that makes a meaningful contribution toward the understanding, prevention, diagnosis, prognosis, treatment and /or rehabilitation of a disease, injury or condition, or to improve the quality of life. Examples include:
  - *data or databases;*
  - *physical collections;*
  - *audio or video products;*
  - *software;*
  - *models;*
  - *educational aids or curricula;*
  - *instruments or equipment;*
  - *research material (e.g., Germplasm; cell lines, DNA probes, animal models);*
  - *clinical interventions;*
  - *new business creation; and*
  - *other.*

Nothing to Report

7. PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

What individuals have worked on the project?
Provide the following information for: (1) PDs/PIs; and (2) each person who has worked at least one person month per year on the project during the reporting period, regardless of the source of compensation (a person month equals approximately 160 hours of effort). If information is unchanged from a previous submission, provide the name only and indicate "no change".

Name:	*Patty Lee, MD*
Project Role:	*Principal Investigator*
Researcher Identifier (e.g. ORCID ID):	*n/a*
Nearest person month worked:	*4*

Contribution to Project: *Dr. Lee supervised the overall design, experimental planning and data interpretation for all the studies.*

Funding Support: *N/A*

Name: *Richard Bucala, MD, PhD*
Project Role: *Co-Investigator*
Researcher Identifier (e.g. ORCID ID): *n/a*
Nearest person month worked: *3*

Contribution to Project: *Dr. Bucala supervised the design and delivery methodology for MIF20.*

Funding Support: *N/A*

Name: *Santos De Jesus Bermejo*
Project Role: *Postgraduate Associate*
Researcher Identifier (e.g. ORCID ID): *n/a*
Nearest person month worked: *4*

Contribution to Project: *Mr. Bermejo assisted with biologic sample collections and maintained all human-related protocols for the Lee lab.*

Funding Support: *N/A*

Name: *Edward Doherty*
Project Role: *Postdoctoral Associate*
Researcher Identifier (e.g. ORCID ID): *n/a*
Nearest person month worked: *10*

Contribution to Project: *Dr. Doherty created MIF20 formulations and assisted with MIF20 delivery to mice.*

Funding Support: *N/A*

Name: *Cheol Hwangbo*
Project Role: *Postdoctoral Associate*

Researcher Identifier (e.g. ORCID ID): *n/a*
Nearest person month worked: *10*

Contribution to Project: *Dr. Hwangbo worked with Dr. Dougherty in all MIF20 delivery mouse experiments, in addition to daily monitoring of the mice, mouse sacrificing and conducting lung assays.*

Funding Support: *N/A*

Name: *So-Jin Kim*
Project Role: *Postdoctoral Associate*
Researcher Identifier (e.g. ORCID ID): *n/a*
Nearest person month worked: *4*

Contribution to Project: *Dr. Kim was trained by Dr. Hwangbo and upon his departure, performed his duties (described above)*

Funding Support: *N/A*

Name: *Nicole Lessard*
Project Role: *Postgraduate Associate*
Researcher Identifier (e.g. ORCID ID): *n/a*
Nearest person month worked: *7*

Contribution to Project: *Ms. Lessard maintained the mouse protocols and renewals, purchased mice, performed acclimation studies before MIF20 and cigarette smoke exposures and maintained the Teague smoking apparatus.*

Funding Support: *N/A*

Name: *Peiying Shan, MD*
Project Role: *Research Associate*
Researcher Identifier (e.g. ORCID ID): *n/a*
Nearest person month worked: *7*

Contribution to Project: *Dr. Shan assisted in daily mouse monitoring, tissue processing and lung function testing of the mice.*

Funding Support: *N/A*

Name: *Yi Zhang*
Project Role: *Associate Research Scientist*
Researcher Identifier (e.g. ORCID ID): *n/a*
Nearest person month worked: *3*

Contribution to Project: *Dr. Zhang assisted Dr. Hwangbo in performing lung histologic analyses and after his departure, continues to train Dr. So-Jin Kim in mouse lung assays.*

Funding Support: *N/A*

Has there been a change in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

If the active support has changed for the PD/PI(s) or senior/key personnel, then describe what the change has been. Changes may occur, for example, if a previously active grant has closed and/or if a previously pending grant is now active. Annotate this information so it is clear what has changed from the previous submission. Submission of other support information is not necessary for pending changes or for changes in the level of effort for active support reported previously. The awarding agency may require prior written approval if a change in active other support significantly impacts the effort on the project that is the subject of the project report.

Two new grants have been awarded and are now active:

R01HL138396 (Lee) 8/15/17 – 5/31/21 2.4 calendar months
NIH/NHLBI $250,000 (annual direct)
TLR4-mediated Epigenetic & Senescence Mechanisms in Emphysema
We will identify TLR4-mediated regulation of cellular senescence via p16INK4a and HDAC2 in emphysema.

VA/ORD #11858595 (Lee) 10/1/16 – 9/30/20 4.0 calendar months
MIF-Mediated Mechanisms in Emphysema $150,000 (year 2 direct)
Goals: This proposal will allow us to define the mechanisms of action of MIF-CD74 and to test the efficacy of targeting MIF-CD74 in the prevention and treatment of emphysema.

What other organizations were involved as partners?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe partner organizations – academic institutions, other nonprofits, industrial or commercial firms, state or local governments, schools or school systems, or other organizations

(foreign or domestic) – that were involved with the project. Partner organizations may have provided financial or in-kind support, supplied facilities or equipment, collaborated in the research, exchanged personnel, or otherwise contributed.

Provide the following information for each partnership:
Organization Name:
Location of Organization: (if foreign location list country)
Partner's contribution to the project (identify one or more)

- *Financial support;*
- *In-kind support (e.g., partner makes software, computers, equipment, etc., available to project staff);*
- *Facilities (e.g., project staff use the partner's facilities for project activities);*
- *Collaboration (e.g., partner's staff work with project staff on the project);*
- *Personnel exchanges (e.g., project staff and/or partner's staff use each other's facilities, work at each other's site); and*
- *Other.*

Nothing to Report

8. SPECIAL REPORTING REQUIREMENTS

COLLABORATIVE AWARDS: N/A

QUAD CHARTS: N/A

9. APPENDICES: N/A