Spring 5-2019

Relationships among Optimism, Inflammation, and Stroke Recovery

Yun-Ju Lai

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RELATIONSHIPS AMONG OPTIMISM, INFLAMMATION, AND STROKE RECOVERY

A DISSERTATION

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR

THE DEGREE OF DOCTOR OF PHILOSOPHY IN NURSING

THE UNIVERSITY OF TEXAS HEALTH SCIENCE CENTER AT HOUSTON

CIZIK SCHOOL OF NURSING

BY

YUN-JU LAI, Ph.D.(c), M.S., R.N.

MAY, 2019
To the Dean for the School of Nursing:

I am submitting a dissertation written by Yun-Ju Lai and entitled "Relationships Among Optimism, Inflammation, and Stroke Recovery." I have examined the final copy of this dissertation for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Doctor of Philosophy in Nursing.

Rebecca Casarez
Committee Chair

We have read this dissertation and recommend its acceptance:

[Signatures]

Accepted
Dean for the School of Nursing
Acknowledgments

I would like to express my deepest appreciation to Dr. Rebecca Casarez, my Committee Chair, and Dr. Geri Wood, for their extraordinary support throughout my dissertation process. They have provided guidance on this project. Moreover, I would like to acknowledge Dr. Louise D. McCullough for her vision, encouragement, and support. She is an exemplary and visionary mentor, a great leader who has dedicated her life to the service of education and healthcare. She is someone I strive to emulate. I would also like to express gratitude to Dr. Liang Zhu for her immense help with my statistical analysis.
Abstract

Yun-Ju Lai, Ph.D.(c), M.S., R.N.

Relationships among Optimism, Inflammation, and Stroke Recovery

May, 2019

Objective: Post-stroke inflammation is detrimental to the brain and results in an unfavorable recovery. Optimism has been associated with lower inflammation and better health outcomes among people with medical conditions, but no studies have assessed this association in the stroke population. The overall goals were to examine the relationships among optimism, stroke severity, physical disability, and inflammation during hospitalization and evaluate the relationships among optimism, inflammation, and stroke recovery over the three-month post-stroke period.

Methods: This study was a secondary analysis of data prospectively collected from the BioRepository of Neurological Diseases biobank. Outcomes included optimism, stroke severity, physical recovery, and inflammatory markers (IL-6, TNF-α, and CRP). Spearman’s correlation, Wilcoxon signed-rank test, multiple linear regression, and mixed-effect regression model were used to determine the relationships among the variables.

Results: A total of 49 subjects at baseline, with 13 at 3-month follow-up were recruited. The results indicated that subjects with higher optimism showed less stroke impairment and lower level of CRP at baseline compared to those with lower optimism. Additionally, optimism was associated with less stroke severity and lower IL-6 and CRP levels over the first three months after stroke.
**Conclusion:** Optimistic stroke survivors showed lower inflammation and better stroke recovery. By understanding this relationship may provide a scientific framework whereby new strategies for stroke recovery can be developed in the future.

**Keywords:** optimism, inflammation, stroke recovery
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Summary of Study

The dissertation study entitled “Relationships among Optimism, Inflammation, and Stroke Recovery” is presented in this book. The purpose of the study was to examine the relationships among optimism, stroke severity, physical disability, and inflammation during hospitalization and evaluate the relationships among optimism, inflammation, and stroke recovery over the three-month post-stroke period. Approval for this study was sought from the Committee for the Protection of Human Subjects (CPHS) at the University of Texas Health Science Center at Houston’s Institutional Review Board and approval was attained in June, 2018.

This book includes (A) a proposal of the study which described study specific aims, background and significance, research design and methods, and research participant risk and protection; (B) a manuscript entitled “Blood Biomarkers for Physical Recovery in Ischemic Stroke: A Systematic Review” which identified 34 biomarkers were significantly associated with physical recovery after ischemic stroke: (1) immune response (15, 44%); (2) lipids/metabolism (4, 12%); (3) neuronal function (4, 12%); and (4) blood vessel/circulation (11, 32%). Of the predictive biomarkers associated with 1-month recovery, 60% (6 of 10) was classified into blood vessel/circulation; 54% (14 of 26) of the biomarkers associated with 3-6 month physical recovery involved the immune response; and (C) a manuscript entitled “Relationships among Optimism, Inflammation, and Stroke Recovery” which found stroke survivors with higher optimism showed less stroke impairment and lower level of CRP at baseline compared to those with lower optimism. Additionally, optimism was associated with less stroke severity and lower IL-6 and CRP levels over the first three months after
stroke. A total of 49 subjects at baseline, with 13 at 3-month follow-up were recruited in this study.

The appendixes contain the Institutional Review Board (IRB) approvals, instruments, and the demographic datasheet used for data collection. The final section of the dissertation is the curriculum vitae of the researcher.
RELATIONSHIPS AMONG OPTIMISM, INFLAMMATION, AND STROKE RECOVERY

A DISSERTATION PROPOSAL

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR

THE DEGREE OF DOCTOR OF PHILOSOPHY IN NURSING

THE UNIVERSITY OF TEXAS HEALTH SCIENCE CENTER AT HOUSTON

CIZIK SCHOOL OF NURSING

BY

YUN-JU LAI, Ph.D.(c), M.S., R.N.

MAY 17, 2018
Abstract

**Background:** Post-stroke inflammation is detrimental to the brain and results in an unfavorable recovery. Optimism has been associated with lower inflammation and better health outcomes among people with cardiac or Parkinson’s diseases, but there are currently no such studies that have been reported in post-stroke population. **Objectives:** The overall objectives of this study are to (1) examine the relationships between optimism, stroke severity, physical disability, and inflammation during the time of hospitalization, and (2) examine the longitudinal relationships between optimism, inflammation, and stroke recovery over the three-month post-stroke period. The long-term goal is to develop a suitable psychological intervention strategy to boost stroke survivors’ optimism levels which may accelerate stroke recovery and reduce risk of recurrent strokes in the future. **Methods:** This study will be a secondary analysis of data previously collected from the BioRepository of Neurological Diseases biobank, utilizing an observational quantitative design. Levels of optimism, stroke severity, physical disability, and plasma inflammatory cytokines, including IL-6, TNFα, and CRP will be measured during the time of hospitalization (T1) and three months following stroke onset (T2). Under the first aim, the correlations among optimism level and stroke severity, physical disability, and levels of inflammatory markers at T1 will be analyzed by Pearson or Spearman correlation coefficient. For the second aim, the correlations of optimism with outcomes of stroke recovery and inflammation will be tested by longitudinal analysis. **Expected outcomes:** The central hypothesis is that patients with higher levels of optimism will show less stroke severity, less physical disability, decreased inflammation, and better physical recovery during the first three months after stroke.
compared to those with lower levels of optimism. **Significance and innovation:** The proposed research is significant because it is expected to provide the better understanding of mechanisms underlying the role of optimism in post-stroke inflammation and physical recovery. The research proposed in this application is innovative, in the applicant’s opinion, because it first (i) examines stability of optimism levels in stroke population by longitudinal analysis; (ii) explains association of optimism with acute and chronic inflammation following stroke; and (iii) explains relationships between optimism, stroke severity, physical disability, and stroke recovery. The rationale for this project is that understanding the relationship between optimism and stroke recovery and the associated mechanism is likely to provide a scientific framework whereby new strategies for stroke recovery can be developed in the future.
Specific Aims

To date, stroke is the fifth cause of death and the most common cause of long-term disability in the United States (Benjamin et al., 2018). Motor impairment, sensory dysfunction, and dysphasia are common symptoms of stroke. Up to 30% of stroke survivors suffer permanent disability, and 20% of survivors required inpatient rehabilitation within three months after a stroke event (Apostolopoulou, Michalakis, Miras, Hatzitolios, & Savopoulos, 2012). In addition, restriction of blood supply to the brain results in decreased delivery of oxygen and nutrients, potentially leading to a permanent brain injury. Inflammatory response induced by damaged and dead cells following stroke may be more detrimental to the brain (Iadecola & Anrather, 2011; Wang, Tang, & Yenari, 2007) and result in an unfavorable recovery (Dziedzic, 2015; J. Y. Kim, Kawabori, & Yenari, 2014).

Optimism is defined as a mood or attitude related to positive expectations of the future and is considered to be a personality trait (Scheier, Carver, & Bridges, 1994; Tiger, 1979). It has been associated with health promotion and disease prevention, such as improvement of surgical recovery (Tindle et al., 2012), post-stroke recovery (Shifren & Anzaldi, 2018a), and decreased the risk of stroke (E. S. Kim, Park, & Peterson, 2011). The previous study also found that optimism level may be significantly increased by various psychological interventions and thereby enhance well-being (Antoni et al., 2001). Accumulating evidence indicates that optimistic people have better mental and physical health (Gison, Rizza, Bonassi, Donati, & Giaquinto, 2015). However, the mechanisms implicated in the links between optimism, inflammation, and post-stroke recovery remain unclear. Therefore, there is a critical need to examine the relationships between
optimism, inflammation, and physical recovery in the post-stroke population. Without such information, the understanding of the connection between mind and body will likely remain limited.

The long-term goal is to develop a suitable intervention strategy to boost stroke survivors’ optimism levels which will accelerate stroke recovery and reduce the risk of recurrent strokes in the future. The overall objectives of this study are to (i) examine the relationships between optimism, stroke severity, and inflammation at stroke onset, and (ii) examine the longitudinal relationships between optimism, inflammation, and physical recovery over three months from stroke onset. The central hypothesis is that patients with higher levels of optimism will show less stroke severity, less physical disability, decreased inflammation, and better physical recovery during the first three months after stroke compared to those with lower levels of optimism. The success of this study may delineate the relationships between optimism and post-stroke recovery, as well as identify the potential inflammatory markers which may predict the prognosis for recovery from stroke. The rationale for this project is that understanding the relationship between optimism and stroke recovery and the associated mechanism is likely to provide a scientific framework whereby new strategies for stroke recovery can be developed.
To attain the overall objectives, we propose a pilot longitudinal study in stroke patients. Assessments of optimism, stroke severity, physical disability, and plasma inflammatory cytokines (IL-6, TNFα, and CRP) will be done during the time of hospitalization (T1) and three months post-stroke (T2). The following two specific aims will be pursued (Figure 1):

**Aim 1: Examine the relationships between optimism, stroke severity, physical disability, and inflammation during the time of hospitalization.**

*Working hypothesis:* stroke patients with higher levels of optimism during the time of hospitalization will show less stroke severity, less physical disability, and lower levels of pro-inflammatory cytokines compared to those with lower optimism levels.

**Aim 2: Examine the longitudinal relationships between optimism, inflammation, and stroke recovery at three-month post-stroke period.**

*Working hypothesis:* patients’ levels of optimism will remain stable over the three months, and more optimistic patients will develop less inflammation and accelerated stroke recovery over the 3-month post-stroke period.

The expected findings of this study provide the critical information on relationships between optimism, inflammation, and stroke recovery. Particular concern was given to boost levels of optimism on physical health among stroke population. The knowledge gained from this research may provide a positive impact on clinical practice.
and be used to design and test an intervention to augment stroke survivors’ optimism level which can improve post-stroke recovery.

**Background and Significance**

According to the recent study by Benjamin et al. (2018), approximately 795,000 Americans have strokes every year. Stroke is the most common neurological disease in the adult population worldwide, and the third cause of chronic disability (Feigin, Norrving, & Mensah, 2017). Motor impairment, sensory dysfunction, and aphasia are common symptoms of stroke. About 25 - 74% of stroke survivors are dependent in activities of daily living (ADL) (Miller et al., 2010). Post-stroke rehabilitation is a continuum, starting within days of stroke onset and ending only when it no longer produces any positive effect. Prior studies indicated that 48 - 91% of physical recovery occurred within the first three months after stroke (Lee et al., 2015; Stinear, 2017; Wade, Wood, & Hewer, 1985). To date, improvements in acute stroke care have reduced stroke-related mortality over the past two decades. However, because of the increased survival rate, many of the survivors are left with severe disability, placing a tremendous burden on the healthcare system and caregivers (F. Mu et al., 2017). Therefore, accelerating physical recovery and preventing of recurrent stroke have become new challenges that await exploration.

Damaged cells caused by stroke may evoke a massive upregulation of the inflammatory response (Dirnagl, Iadecola, & Moskowitz, 1999). Pro-inflammatory cytokines, such as IL-6, TNFα or CRP, attract innate immune cells from circulatory system to infiltrate the damaged tissue and support post-stroke healing (Lakhan, Kirchgessner, & Hofer, 2009; Simats, Garcia-Berrocoso, & Montaner, 2016). The
infiltrated immune cells may produce cytotoxic mediators that prolong the inflammatory response, increase brain damage and contribute to edema formation and hemorrhagic transformation (Hu et al., 2016). These secondary complications commonly influence stroke outcomes and leave an individual with residual impairment of physical, psychological, and cognitive functions. Hence, properly regulating the post-stroke inflammation may be a therapeutic strategy to reduce the secondary brain injury and enhance post-stroke recovery (Simats et al., 2016).

Optimism is conceptually defined as positive expectations of the future (Scheier & Carver, 1985). It is considered a personality trait, but also a positive psychological factor (Conversano et al., 2010b; Scheier & Carver, 1985). Studies have shown that optimistic expectation may change over time whenever a person’s situation changes, such as in high-stress situations (Atienza, Stephens, & Townsend, 2004; Segerstrom, 2007). A meta-analysis indicated that psychological intervention approaches, such as Best Possible Self or cognitive-behavior therapy may be able to significantly augment level of optimism among different populations (Malouff & Schutte, 2017).

Optimistic individuals tend to be positive about events happened in their daily life. Accumulated studies have found that optimism is associated with beneficial health outcomes. For example, optimistic people report more resilience to stress, faster recovery after cardiac bypass surgery (Ronaldson et al., 2014; Tindle et al., 2012), less chronic inflammation in older adults (Ikeda et al., 2011) and lower blood pro-inflammatory cytokines in patients with cardiovascular diseases (Roy et al., 2010). Therefore, optimism is not only critical to maintaining healthy state mentally, but it may also improve the therapeutic outcome physically. Most of stroke survivors suffer from the physical
disability. A qualitative study found that optimistic stroke survivors may exhibit more confidence to face the challenges of stroke recovery (Jones, Mandy, & Partridge, 2008). Moreover, a recent study also showed that stroke survivors with higher levels of optimism develop fewer depressive symptoms and better perceived physical health during rehabilitation (Shifren & Anzaldi, 2018a). However, no studies have examined the links between optimism, inflammation, and physical recovery among stroke population.

The purpose of this study is to examine the relationships between optimism, inflammation, and physical recovery among stroke population. Given the evidence of associations of optimism, inflammation, and physical health with multiple diseases, but not stroke, it is important to determine how optimism is associated with inflammation and post-stroke recovery. With the understanding of mechanisms underlying the role of optimism in post-stroke inflammation and physical recovery, psychological interventions for boosting optimism may be developed.

**Conceptual Framework**

To examine the relationships between optimism, inflammation, and post-stroke recovery, the conceptual framework will adapt from Kang’s biobehavioral model (Kang, Rice, Park, Turner-Henson, & Downs, 2010). It includes (1) psychosocial factors, such as stress or optimism, represents psychological and social factors that influence health; (2) biological factors, such as immune or endocrine functions, are defined as body’s physiological reactions; and (3) health outcomes are a change in the health status of an individual, group or population. The model can explore the relationship between psychosocial factor (optimism), biological factor (inflammation), and health outcomes (physical disability and recovery) in stroke patients (Figure 2). The biobehavioral model
contributes to the understanding of health and health-related outcomes and may serve as a basis for developing and evaluating appropriate interventions such as cognitive behavioral therapies to promote, restore, and maintain health. Through this model, we can have a better understanding of the interaction between mind and body among post-stroke population. The successful completion of the proposed study will provide the critical evidence to link optimism, inflammation, and physical recovery in the stroke population.

**Innovation**

Optimism has been associated with decreased inflammatory response among people with cardiovascular diseases (Roy et al., 2010) and better functional outcomes and quality of life in patients with Parkinson’s disease (Gison et al., 2015) or traumatic brain injuries (Ramanathan, Wardecker, Slocomb, & Hillary, 2011). However, there are no studies that have examined the links between optimism and inflammation in stroke recovery. As such, we believe this study is innovative because it first:

1. Examines stability of optimism levels in stroke population by longitudinal analysis.
2. Explains association of optimism with acute and chronic inflammation following stroke.

*Figure 2. The conceptual model. Conceptual framework of psychosocial factor, physiological factor, and health outcomes post-stroke population is adapted from Kang’s biobehavioral model.*
3. Explains relationships between optimism, stroke severity, physical disability, and stroke recovery.

In this regard, understand the association of optimism in inflammation and post-stroke recovery may serve as a basis for developing and evaluating appropriate psychological interventions to boost optimism to accelerate stroke recovery in the future.

**Research Design and Methods**

To understand how optimism influences stroke severity, inflammation, and recovery, we will test the working hypotheses of Aim 1 and Aim 2. Minimal evidence regarding the relationships between these variables at stroke onset and 3-month following stroke. Filling this gap is likely to improve the understanding of the role of optimism in physical function, stroke recovery and inflammation over a three-month period. Upon completion of this study, less stroke severity, decreased physical disability, lower inflammation, and better recovery will be expected in more optimistic stroke survivors.

**Research Design**

This study will be a secondary analysis of data previously collected from the BioRepository of Neurological Diseases biobank, utilizing an observational quantitative design. This study will investigate how optimism influences (1) stroke severity, physical disability, and inflammation during the time of hospitalization (T1) in Aim 1; (2) stroke

![Figure 3. Timepoints of sample collection. LOT-R indicates Revised Life Orientation Test; NIHSS indicates National Institutes of Health Stroke Scale; mRS indicates modified Rankin Scale.](image)
recovery and inflammation over a three-month post-stroke period (T1 and T2) in Aim2 (Figure 3).

**Sample and Setting**

**Inclusion criteria** in this study are: the participant is (1) aged ≥ 18 years; (2) diagnosed with an ischemic stroke.

**Exclusion criteria** are: the participant is (1) diagnosed with transient ischemic attack or intracerebral hemorrhage, including secondary hemorrhage on control computed tomography or magnetic resonance imaging (to avoid confounding effects of hematoma-induced inflammation (2) with underlying vascular lesions or traumatic brain injury, systemic malignancy, autoimmune disease, or immunosuppression (defined as current (>28 day) use of any immunosuppressive drugs; and (3) unable to provide informed consent will be excluded.

For Aim 1, we assume that the minimum correlation coefficient between optimism and stroke severity, physical disability, and inflammation is 0.4 (Ikeda et al., 2011; Roy et al., 2010). To identify a correlation coefficient with 80% of power at a type I error of 0.05, we need 46 participants (Bujang & Baharum, 2016). For Aim 2, we need to compare the outcomes of stroke recovery and inflammation between optimism and non-optimism groups. Based on related articles and our preliminary data, we assume that the mean differences are 8.2 with a standard deviation (SD) of 8.1 for NIHSS, 2.3 with a SD of 1.7 for mRS, and 1.8 with a SD of 2.2 for CRP. The required sample sizes for the three variables are 24, 14, and 36, respectively, to achieve 80% of power at a type I error of 0.05 using longitudinal data analysis (Hedeker, Gibbons, & Waternaux, 1999). The maximum sample size among the three variables is 40 even after Bonferroni correction
for multiple testing. As a result, the sample size 46 from Aim 1 is sufficient for Aim 2 as well. Additionally, with an estimated dropout rate of 15%, 54 participants need to be recruited in this study.

**Data Collection**

The data will be obtained from the BioRepository of Neurological Diseases biobank. The data, including optimism, stroke severity, and physical disability will have previously been gathered at two time points, (1) T1: during the time of hospitalization and (2) T2: three months, with windows of ±14 days, after stroke onset. Participant’s demographic and clinical data, such as age, gender, race/ethnicity, and medical histories will be extracted from the biobank (Figure 3). Peripheral venous blood samples will have been obtained from participants within 24±6 hr from admission (T1) and three months ±14 days after stroke onset (T2).

**Measurements**

The main predictor variable is level of optimism which will be measured by LOT-R (Scheier et al., 1994). The range of scores is from zero to 24, with higher scores indicating higher levels of optimism. It includes 10 items; four of them are filler items which are not used in scoring. Of the six items that are scored, three are keyed in a positive direction and three in a negative direction. The respondents indicate the extent to which they agree with each item on a 5-point Likert scale that ranges from strongly disagree to strongly agree. The acceptable internal consistency of LOT-R was reported as Cronbach’s α of 0.82 (Shifren & Anzaldi, 2018a) in stroke population and stability (test-retest reliability) over a 4-month period (r = 0.79) in college students (Scheier et al., 1994). For indicative purposes, optimism level will use cutoff values as follows: 0-13
indicates low optimism, 14-18 moderate optimism and 19-24 high optimism (Kreis et al., 2015).

The outcome variables are (1) Severity of stroke will be evaluated by the National Institutes of Health Stroke Scale (NIHSS) by the certified neurologist at T1 and T2 (Brott et al., 1989). The data will be extracted from the biobank at T1 and T2. The range of scores is from zero to 42, with lower scores indicating less impairment. The acceptable interrater reliability was reported as an intraclass correlation coefficient of 0.82 (Goldstein & Samsa, 1997) and high content validity in stroke population (Kasner, 2006a). (2) Physical disability after a stroke will be assessed with modified Rankin Scale (mRS) by the certified neurologist at T1 and T2 (Banks & Marotta, 2007). The data will be extracted from the biobank at T1 and T2. The range of mRS scores is from zero to six, with lower scores indicating less disability. The acceptable internal consistency of mRS was reported as Cronbach’s α of 0.89 in stroke survivors (Wei, Han, Wei, & Duan, 2015). (3) IL-6, TNF-α, and CRP are defined as plasma inflammatory biomarkers. Levels of biomarkers will be measured at T1 and T2 by multiplex ELISA. The assays have a lower limit of sensitivity (minimum detectable concentration) of 1-2 pg/ml, and the average intra- and inter-assay coefficients of variations reported by the manufacturer are <10% and <10%, respectively.

Blood sample will be collected as follows: Immediately after blood samples are collected, participants will be de-identified and assigned a unique subject number. Blood samples will be labeled with preprinted barcode labels. Plasma will be separated by centrifuging at 1,000-2,000xg for 10 minutes. Following centrifugation, plasma will be immediately transferred into a clean collection tube and maintained at 2-8°C while
handling. To avoid freeze-thaw cycle, plasma will be separated into aliquots at a volume of 0.1ml. Plasma samples will be analyzed for inflammatory markers, including IL-6, TNF-α, and CRP by multiplex enzyme-linked immunosorbent assay (ELISA) (BioRad, Hercules, CA) in the end of each month. All samples will be analyzed by a researcher blinded to participant’s stroke severity (NIHSS) and physical disability (mRS). Rest of plasma will be stored in -80°C freezer for future study. Freezers for storage will be located on the 3rd floor of the UTHealth McGovern Medical School MSE R319 (Dr. McCullough’s laboratory).

Potential confounder variables including age, gender, history of stroke, stroke severity, and medical histories, medicine records, and social support will also be examined in both Aim 1 and Aim 2.

**Statistical Procedures**

Following a test of statistical normality, descriptive statistics, including the number of participants, age, gender, and race/ethnicity will be expressed as mean ± SD or median with interquartile range. To demonstrate the hypothesis of Aim 1, we will calculate the Pearson or Spearman correlation coefficient among optimism score and stroke severity, physical disability, and levels of inflammation biomarkers at T1 using SPSS 25.0 for Windows (SPSS Inc., Chicago, IL). The correlation among variables and potential confounders will be analyzed by multiple linear regression. Values of p < 0.05 will be considered statistically significant.

To validate the hypothesis of Aim 2, we will compare the outcomes of stroke recovery and inflammation between optimism and non-optimism groups by longitudinal analysis, i.e., linear mixed model, including time, group, and their interactions in the
model. In addition, we will use continuous optimism score in the longitudinal analysis for further details. Bonferroni correction will be used to adjust for the multiple testing. For missing data, if missing is random, we will analyze the data as it is. Otherwise, we will follow Ibrahim and Molenberghs’ method for data analysis (Ibrahim & Molenberghs, 2009). Values of \( p < 0.05 \) will be considered statistically significant.

**Data Management**

All study data will be entered into a secure online database created and managed by UT Houston Data Management and Analysis Core (DMAC) at the Department of Neurology / Stroke Institute. The database will be created using the Research Electronic Data Capture (REDCap) database management system. Data will be stored on secure zone 100 UTHealth servers which are managed by UTHealth School of Bioinformatics and have been approved for collection and storage of patient information. Access to the database will follow all security protocols of UTHealth Houston. Access will be managed centrally via the DMAC and will be assigned based on personnel roles on the principle of least privilege (i.e. access only to the information and resources that are necessary for individuals’ legitimate purpose and function). All personnel obtaining access to the database will undergo individual identification verification and management system of the UTHealth Houston. Other than assigned DMAC personnel, no users will have the capacity to download raw data. Within the DMAC no downloaded data with PHI will be stored on local computers or removable media. Such data will be stored and managed on UTHealth Neurology Servers (NAS). Data for analyses will be generated by DMAC using the de-identified IDs.
Strengths of the Proposed Study

The proposed study is unique in being the first study of the relationships between optimism, inflammation, stroke severity, and functional recovery in post-stroke population. Detection of inflammatory biomarkers in blood at the time of admission and 3-month after stroke onset can provide valuable information regarding the pathophysiology of disease and predict patient recovery better than standard clinical prediction tools (Misra et al., 2017; Unden et al., 2009). Furthermore, the success of this study may help design a proper psychological intervention to boost the level of optimism.

Research Subject Risk and Protection

Participants may involve potential risks of a breach of confidentiality. The risks will be minimized by (1) Removing direct participant identifiers (i.e., names, medical record numbers) from samples and associated data stored in the research repository. All participants will be assigned a unique subject number (de-identified). All samples will be labeled with preprinted barcode labels using the same numbering system. (2) Limiting access to samples and associated data contained within the research repository to the research team; (3) Securing in a separate location, and limiting access to information linking codes (i.e., linkage codes) assigned to the repository samples and associated data with direct participant identifiers.

Potential Pitfalls & Alternative Strategies

The main potential pitfall is loss of confidentiality of the data. This risk will be minimized as above under Potential Risks and Controls. Moreover, because the cytokine analysis may involve more complicated analysis, we will be assisted by Dr. Liang Zhu in the statistical analysis at the UTHHealth McGovern Medical School. In the meantime, we
will routinely consult with Dr. Rebecca Casarez at the UTHealth Cizik School of Nursing and Dr. Louise McCullough at the Department of Neurology, UTHealth McGovern Medical School. They will supervise the project and provide professional opinions.

**Ethical Aspects of the Proposed Research**

Participant’s medical condition will be managed according to the guidelines published by the American Stroke Association (Powers et al., 2018). This study will be approved by the UTHealth CPHS.

**Timeline of the Proposed Research**

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| Recruitment | X | X | X | X | X |
| Data Collection | X | X | X | X | X | X | X |
| Data analysis: Aim 1 |     | X | X | X |
| Data analysis: Aim 2 |     | X | X | X |
| Report preparation | X | X | X |
| Dissemination: dissertation |     | X | X |
References


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Multi-Ethnic Study of Atherosclerosis (MESA). *Psychosomatic Medicine, 72*(2), 134-140. doi:10.1097/PSY.0b013e3181cb981b


March. 27, 2019

Editorial Board
American Journal of Translational Research

Dear Editor,

Enclosed please find our manuscript entitled “Blood Biomarkers for Physical Recovery in Ischemic Stroke: A Systematic Review” which we would like to submit for consideration for publication as a Review Article in American Journal of Translational Research.

Blood biomarkers have been used to predict prognosis in ischemic stroke, but studies linking blood biomarkers to physical recovery after ischemic stroke have not been systematically reviewed since 2011. In this manuscript, we analyzed the articles reported between January 2011, and September 2018 from PubMed, Embase, and CINAHL databases. A total of 34 biomarkers were identified. These were further characterized into four broad categories: (1) immune response, (2) lipids/metabolism, (3) neuronal function, and (4) blood vessel/circulation based on the pathological and biological relevance. Our data summarizes the important biomarkers with prognostic information that may assist clinicians with patient-centered rehabilitation efforts.

On behalf of all authors, please find the required documents submitted for publication consideration. All authors have read and approved the submitted manuscript; the manuscript has not been submitted elsewhere nor published elsewhere in whole or in part. The authors have no conflicts of interest to report and have adhered to all authorship and ethical adherence best practices. To facilitate the reviewing process, we would like to recommend the following experts in the field as potential reviewers:

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We greatly appreciate your time and look forward to hearing from you soon.
Yours sincerely,

Yun-Ju Lai, Ph.D. (c), MS, RN

UTHealth | Cizik
School of Nursing
Blood Biomarkers for Physical Recovery in Ischemic Stroke: A Systematic Review

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Disclosures

None
Abstract

Stroke is a common cause of physical disability. Biomarkers have been used to predict prognosis in ischemic stroke, but studies linking biomarkers to physical recovery from ischemic stroke have not been systematically evaluated since 2011. The purpose of this paper is to report the findings of a systematic review of the intervening literature to identify potential predictive biomarkers for recovery of physical function following ischemic stroke. The PubMed, Embase, and CINAHL databases were searched for studies reported between January 1, 2011, and September 18, 2018. Search criteria were adult ischemic stroke patients, blood sample collection within 24±6 hrs of stroke onset, and outcome measures, including physical function. Identified from 18 studies and representing four biological classifications, 34 biomarkers were significantly associated with physical recovery after ischemic stroke: (1) immune response (15, 44%); (2) lipids/metabolism (4, 12%); (3) neuronal function (4, 12%); and (4) blood vessel/circulation (11, 32%). Of the predictive biomarkers associated with 1-month recovery, 60% (6 of 10) was classified into blood vessel/circulation; 54% (14 of 26) of the biomarkers associated with 3-6 month physical recovery involved the immune response. Blood biomarkers might provide useful information to improve the prediction of physical outcome after ischemic stroke. The data suggest that biomarkers from four biological classifications may predict physical recovery in patients after ischemic stroke.

Key words: biomarker, ischemic stroke, recovery of function
Introduction

Stroke is the 5th leading cause of death in the U.S. (Benjamin et al., 2018), with an annual incidence of approximately 795,000 cases. Ischemic stroke accounts for 87% of all strokes (Benjamin et al., 2018). In addition, stroke is the most common neurological disease in the adult population worldwide, and the third leading cause of chronic disability (Feigin et al., 2017). Up to 74% of stroke survivors are dependent in activities of daily living (Miller et al., 2010).

Motor impairment, sensory dysfunction, and dysphasia are common manifestations of stroke. Up to 30% of stroke survivors in the U.S. suffer permanent disability, and 20% of survivors require inpatient rehabilitation within 3 months after stroke (Creutzfeldt, Holloway, & Walker, 2012). Improvements in acute stroke care have reduced stroke-related mortality over the past two decades; however, the increased survival rate leaves many survivors with severe disability, placing a tremendous burden on the healthcare system and caregivers (F. Mu et al., 2017).

Prior studies indicated that up to 91% of physical recovery occurs within the first 3 months after stroke (Lee et al., 2015). Physical recovery includes motor function, sensation, language, and swallowing ability (Harvey, 2015). Because these physical functions often are interdependent, all are included in the assessment of recovery after stroke, and no single measure fully describes disability or functional outcome from stroke.

The most widely used scales for stroke outcomes are the National Institutes of Health Stroke Scale (NIHSS), the modified Rankin Scale (mRS), and the modified Barthel Index (mBI) (Harrison, McArthur, & Quinn, 2013). The NIHSS is used to
quantify stroke severity based on language, motor function, sensory loss, consciousness, visual fields, extraocular movements, coordination, neglect, and speech (Adams et al., 1999). Global disability, with a focus on mobility, is assessed with the mRS (Banks & Marotta, 2007). Functional outcomes and daily life activities are measured with the mBI (Ohura, Hase, Nakajima, & Nakayama, 2017). A statistically significant inverse correlation has been demonstrated between the mRS and the mBI in the post-stroke population (Memis, Kozanoglu, Kelle, & Goncu, 2016); the more disability on the mRS, the less independent the patient scores on the mBI. Furthermore, admission NIHSS score has been found to be positively correlated with mRS score over time (Saver & Altman, 2012).

A biomarker is a molecule measured in blood, urine, cerebrospinal fluid, or tissue, or an imaging test, such as magnetic resonance imaging or computed tomography. Blood biomarkers have been commonly used to provide prognostic information following ischemic stroke. For example, levels of brain natriuretic peptide (BNP), D-Dimer, matrix metallopeptidase-9 (MMP-9), and S100B in blood were positively correlated with mortality at 4 months post-stroke (Iemolo, Sanzaro, Duro, Giordano, & Paciaroni, 2016). Cardiac markers, such as BNP and troponin, have been shown to have a consistent association with poor outcome after ischemic stroke (Whiteley, Chong, Sengupta, & Sandercock, 2009). Moreover, the most recent systematic review of blood biomarkers for acute stroke found that levels of glucose, glutamate, and fibrinogen at stroke onset were associated with poor prognosis in ischemic stroke patients (Hasan, McColgan, Bentley, Edwards, & Sharma, 2012).
Emerging biomarkers from new discoveries in the field of stroke research may help predict stroke outcome and recovery. As the Hasan et al. (Hasan et al., 2012) publication is the most recent and relevant systematic review, the purpose of this review was to synthesize the literature published after 2011 on the relationship between biomarkers and physical recovery from ischemic stroke. Ultimately, biomarkers may provide prognostic information to guide clinicians with patient-centered rehabilitation efforts.

**Methods**

**Search Strategy**

PubMed, Embase, and CINAHL databases were searched for studies that examined blood biomarkers and functional outcome in patients with ischemic stroke. The hierarchical search strategies and keywords were (1) biomarker, (2) ischemic stroke, (3) physical recovery, and (4) adult (Supplementary Table 1). Duplications were removed from the list, and titles and abstracts of articles retrieved were reviewed for eligibility. All relevant articles from reference lists of each reviewed paper were identified. After screening the abstracts, final eligibility was determined based on the full content of the articles.

**Inclusion and Exclusion Criteria**

Inclusion criteria for the selection of articles to include in the review were: (1) primary source of quantitative study in a peer-reviewed journal published in English between January 2011 and September 2018; (2) patients ≥ age 18 years diagnosed with ischemic stroke; (3) biomarkers measured from blood samples within 24±6 hrs of stroke onset; (4) measure(s) of physical outcome (e.g., NIHSS, mRS and/or mBI); and (5)
reported the association with physical outcome for each biomarker. Note that the short window, within 24±6 hrs for blood sampling relative to stroke onset, maximizes the predictive potential of the biomarkers to provide time-sensitive prognostic information in the early clinical evaluation of ischemic stroke. Unpublished theses, dissertations, and conference proceedings were excluded.

Study Quality Appraisal

A code sheet was used to extract information from each article. The information included author(s), year of publication, subjects, time frame of blood draw, biomarker(s), measurement of physical outcomes, and findings (Cooper, 2017, pp. 110-150). Where more than one cohort was examined within a study, the results for each cohort were extracted separately. Quality of the study was assessed using the modified questionnaire implemented by Whiteley et al. (Whiteley et al., 2009) for the stroke population (Supplementary Table 2). These quality components correspond to the sections on study design and assay methods sections of the REporting recommendations for tumor MARKers prognostic studies (REMARK) (McShane et al., 2005).

Results

Based on the search keywords, 333 studies were identified from PubMed, Embase, and CINAHL. Three more studies were found through Google Scholar and reference lists of relevant articles. The search procedure (Fig 1) followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) flow diagram (Liberati et al., 2009). After screening the title and abstract of each article, 98 full-text studies were assessed for eligibility; 18 studies met the eligibility criteria and were included in the systematic review.
Characteristics of the Studies

The articles are listed in ascending chronological order in Supplementary Table 3. Studies in this review were published between 2011 and 2018. Sample sizes varied from 50 to 783 patients. The proportion of male patients was higher than females (54.3\% male vs. 45.7\% female). The overall means of patient ages varied from 59 to 73 years. All studies enrolled patients with ischemic stroke, and 5 (28\%) included healthy controls as clinical comparisons. Only 2 (11\%) of the studies selected patients with first-episode ischemic stroke; the others did not explicitly address history of stroke. Studies were conducted in Spain (7, 39\%), Germany (3, 17\%), Japan (2, 11\%), and 1 (6\%) each in the Netherlands, Turkey, the United Kingdom, Poland, the United States, and Italy. Of 73 molecules tested in the 18 studies, 35 (48\%) were significantly associated with physical recovery after ischemic stroke.

Methodological Assessment

The percentages of reports reviewed that met the modified REMARK criteria for methodological quality are shown in Supplementary Table 4 and Figure 2. All study authors defined clinical outcome and provided the characteristics of the study population; 94\% used a prospective design; 94\% developed a logistic regression model and reported such adjustment variables as age and stroke severity; 72\% defined enrollment period; and 67\% provided information on the measurement of biomarkers. Few authors reported sample size calculation (6\%), and blinded biomarker measurement (33\%).

Physical function was defined as an individual's capacity to perform daily personal living tasks like eating, walking, and bathing (Ilunga Tshiswaka, Seals, & Raghavan, 2018). A number of outcome measures following ischemic stroke were
reported (Supplementary Table 3). Various instruments (NIHSS, mRS, or mBI) were used and outcomes were measured at different time points after stroke (admission, 1, 3, or 6 months). All studies presented NIHSS stroke severity at admission, but only one study reported the 3-month NIHSS score. Therefore, in this review, NIHSS score was not used to define physical recovery from ischemic stroke. In 15 studies (83%), physical outcomes at 3 months were evaluated by mRS or mBI; 2 (11%) and 1 (6%) of the studies reported physical outcomes at 1 and 6 months after stroke, respectively. Poor outcome was defined according to definitions used in the reviewed articles: mRS ≥ 3 in 13 (72%) studies, mRS ≥ 2 in 2 (11%) studies and mBI < 15 in 1 (6%) study; two studies did not provide cut-off scores for the mRS. Likewise, for this review, we defined physical recovery as used by the authors of the reviewed articles.

**Biomarker Findings**

**Biomarkers related to biological functions.** The 34 biomarkers that were significantly associated with physical outcome after ischemic stroke were divided into four categories based on biomarker biological function (Table 1). In terms of immune response, patients with lower levels of C-C motif chemokine 11 (CCL11), interleukin (IL)-1β and IL-8, and monocyte chemoattractant protein 1 (MCP1) and higher levels of adiponectin, IL-1Ra, IL-6, IL-10, IL-12, copeptin, C-reactive protein (CRP), growth differentiation factor-15 (GDF-15), osteopontin, tumor necrosis factor-alpha (TNFα), and white blood cell (WBC) count at stroke onset showed worse physical recovery (poor outcomes at 1, 3, or 6 months post-stroke). Notably, patients with poor outcome exhibited more than twofold higher levels of copeptin, IL-6, and TNFα compared to those with good outcome.
For lipid/metabolism biomarkers, decreased high-density lipoprotein cholesterol (HDL-C) and increased cholesterol, low-density lipoprotein cholesterol (LDL-C), and glucose at stroke onset were significantly associated with worse functional outcome at 1, 3, or 6 months post-stroke. With respect to biomarkers of neuronal function, patients with ischemic stroke who had higher levels of glutamate and lower levels of glutamic oxaloacetic transaminase (GOT), glutamic pyruvic transaminase (GPT), and brain-derived neurotrophic factor (BDNF) tended to have worse outcomes at 1, 3, or 6 months after stroke. Patients with poor outcome at 1, 3, or 6 months post-stroke showed increased levels of the blood vessel/circulation biomarkers D-Dimer, endostatin, fibronectin, pro-matrix metalloproteinase-10 (proMMP-10), troponin, and midregional proatrial natriuretic peptide (MR-proANP) and decreased levels of estimated glomerular filtration rate (eGFR), creatinine clearance (CrCl), hematocrit, and hemoglobin at stroke onset. Furthermore, elevated levels of fibrinogen at 1 and 3 months after stroke were associated with poor outcome.

**Biomarkers related to physical recovery.** Among 34 blood biomarkers that were significantly associated with physical outcome after ischemic stroke (Table 2), 10, 26, and 2 biomarkers, respectively, showed a significant association with physical outcome at 1, 3, or 6 months following ischemic stroke: 60% (6 of 10) of biomarkers predictive of physical outcome at 1 month were classified in the blood vessel/circulation category; 54% (14 of 26) of biomarkers that predicted recovery between 3 and 6 months post-stroke were related to the immune response. Elevated levels of CRP and fibrinogen were found to be significant predictors of poor prognosis at 1 and 3 months post-stroke.
Low level of CCL11 and high level of blood glucose were significantly associated with poor outcome at 3 and 6 months after ischemic stroke. Nine (50%) of the reviewed studies reported biomarkers and physical outcomes in patients who had received tissue plasminogen activator (tPA) intravenously, the gold standard treatment of acute ischemic stroke (Adeoye, Hornung, Khatri, & Kleindorfer, 2011). In this review, patients who received tPA treatment with poor physical outcomes compared to those with good outcomes had higher levels of IL-6, TNFα, osteopontin, fibronectin, endostatin, proMMP-10, CRP, IL-1Ra, IL-10, and IL-12; and lower levels of HDL, BDNF, IL-1β, IL-8, and MCP1.

Discussion

Of 73 putative biomarkers tested, 34 were found in this review to be statistically significantly associated with physical recovery after ischemic stroke. The biomarkers showed an association with poor outcome by different instruments (mRS or mBI) and measurement time points (admission, 1, 3, or 6 months after stroke). To provide a functional view of the biomarkers, we divided the 34 into four categories based on biological function of the individual biomarker: immune response, lipids/metabolism, neuronal function, and blood vessel/circulation.

Immune Response

Brain injury from ischemia is exacerbated by the inflammatory response to cell injury and necrosis (Iadecola & Anrather, 2011; Wang et al., 2007). To repair tissue damage, inflammatory cytokines and chemokines attract immune cells from the circulation into the brain (Lakhan et al., 2009; Simats et al., 2016), and over-activated immune cells adversely augment brain damage leading to an unfavorable recovery
(Dziedzic, 2015; J. Y. Kim et al., 2014). We found that 54% of the 26 biomarkers that predicted long-term (3-6 month) stroke recovery are related to immune response. Consistently, IL-6 (Gori et al., 2017; Rodriguez et al., 2013), TNFα (Gori et al., 2017; Rodriguez et al., 2013), and CRP (De Marchis et al., 2013; De Marchis et al., 2018; Gori et al., 2017) were found to be robust predictors of long-term functional outcome in ischemic stroke. Therefore, higher levels of immune-related biomarkers after ischemic stroke may reflect worse physical recovery.

**Lipids/Metabolism**

We identified four lipid/metabolism biomarkers (high glucose, cholesterol, HDL-C, and low LDL-C) that were significantly associated with poor outcome after ischemic stroke. High blood glucose, cholesterol, and LDL-C and low HDL-C levels have been associated with increased risk for atherosclerosis and stroke (Brites, Martin, Guillias, & Kontush, 2017; Giacco & Brownlee, 2010; Zhao et al., 2016). LDL-C and hemoglobin A1c (HbA1c) levels guide recommendations in the American Heart Association/American Stroke Association stroke prevention guidelines (Kernan et al., 2014). A prior systematic review indicated that low HDL-C level is associated with worse physical outcome after ischemic stroke (Amarenco, Labreuche, & Touboul, 2008). Five of the articles (Brea et al., 2011; Campos et al., 2011; De Marchis et al., 2013; De Marchis et al., 2018; Luitse et al., 2013) reviewed in this paper indicated that hyperglycemia was associated with poor outcome in patients with ischemic stroke, which is consistent with previous studies (Hasan et al., 2012; Nair, Sylaja, Sreedharan, & Sarma, 2017; Putaala et al., 2011). Hasan et al. (Hasan et al., 2012) reported that admission glucose level might predict poor outcome following tPA treatment; however,
the majority of biomarkers we found associated with physical recovery after treatment with tPA were related to immune response and blood vessel/circulation, not to glucose or other biomarkers in the lipid/metabolism category.

**Neuronal Function**

Although different brain regions have different thresholds for ischemic cell damage, neurons are the most sensitive to hypoxia (Woodruff et al., 2011). Glutamate is the major excitatory neurotransmitter in the brain that mediates the signal of neuronal degeneration following ischemic stroke (Bano & Nicotera, 2007; Lai, Zhang, & Wang, 2014). Elevated levels of glutamate, with decreased GOT and GPT (enzymes to decrease the level of glutamate in peripheral blood), may induce neuronal apoptosis (Iadecola & Anrather, 2011; Teichberg, Cohen-Kashi-Malina, Cooper, & Zlotnik, 2009). Higher levels of glutamate, and lower levels of GOT, GPT, and BDNF were associated with less favorable physical recovery after ischemic stroke in this review. Hasan et al. (Hasan et al., 2012) reported similar findings that elevated glutamate may indicate progressive stroke.

**Blood Vessel/Circulation**

Ischemia disrupts the mitochondrial membrane potential, which generates excessive reactive oxygen species (ROS) in endothelial cells of cerebral blood vessels (Kalogeris, Bao, & Korthuis, 2014). ROS damages mitochondrial DNA, activates the inflammatory response, induces secretion of MMPs, and leads to endothelial cell swelling and death (Hu et al., 2016). This process triggers breakdown of the blood-brain barrier and thus increases the risk of hemorrhagic transformation (i.e., bleeding into an area of
ischemic brain when cerebral blood flow is restored to damaged vasculature), which adversely affects stroke outcome.

Fibrinogen, an acute phase protein, is involved in platelet activation, coagulation, and hemostasis (Koenig, 2003). Our results support earlier findings that elevated fibrinogen level is associated with poor functional outcome at 1 (Potpara et al., 2014) and 3 months (Campos et al., 2011) after ischemic stroke. Interestingly, a prior study indicated that an early reduction in fibrinogen increases the risk of intracerebral hemorrhage after tPA treatment in ischemic stroke patients (Vandelli et al., 2015). Therefore, the relationship between fibrinogen and physical outcome among ischemic stroke patients, with or without tPA intervention, merits further research.

**Strengths and Weaknesses of the Research**

A strength of this systematic review is the criterion for timing of blood collection for biomarker determination. The short window – within 24±6 hrs – for blood sampling relative to stroke onset maximizes the predictive potential of the biomarkers to provide time-sensitive prognostic information in the early clinical evaluation of ischemic stroke. Moreover, because stroke recovery is a chronic process, this review addressed potential prognostic prediction of physical recovery at 1, 3, and 6 months after ischemic stroke. Categorization of predictive biomarkers into four broad categories based on biological function may further inform our understanding of the clinicopathology of stroke. On the other hand, the short window for blood collection precluded biomarkers that are expressed in the later phases of ischemic stroke, which may have prognostic importance for physical recovery. Congruent with the prior systematic review (Hasan et al., 2012),
high levels of glucose, glutamate, and fibrinogen within 24±6 hrs of stroke onset were repeatedly found to be associated with poor outcome after ischemic stroke.

The review is limited by the information that could not be accounted for because it was either not controlled or not reported in the primary source articles. Biomarker expression typically follows a circadian rhythm. For example, expression of TNFα, IL-1β, and IL-6 is known to peak in the evening (Scheiermann, Kunisaki, & Frenette, 2013), whereas copeptin level remains unchanged throughout the 24-hour day (Beglinger, Drewe, & Christ-Crain, 2017). Unknown biomarker expression pattern and/or inconsistency in timing of blood collection within and across studies could have adversely affected the findings. Such issues should be addressed in future prospective studies on biomarker expression after ischemic stroke. Another limitation is the small sample size in some of the eligible studies, which inherently compromises the statistical power of the relationships between blood biomarkers and physical recovery from ischemic stroke.

**Implications for Clinical Practice and Future Research**

The biomarkers we found in the acute period may provide useful insights into prognosis and mechanism(s) of action. These findings can facilitate biomarker-based care in patients with ischemic stroke, and thus match patients with appropriate intervention. A robust number and types of biomarkers have been and continue to be investigated in patients with ischemic stroke. Biomarkers show promise, especially in the prognosis of functional recovery and evaluation of therapeutic response, but the current application of their usefulness in practice is elusive without further research evidence from controlled studies. Future research is expected to align timing of putative predictive biomarker
levels with phases of physical recovery; the timing may well correlate with the biological classification of the biomarker and suggest different pathways that are mechanistically different for acute and chronic stroke recovery. For instance, troponin, a blood vessel/circulation marker, indicates myocardial damage in acute coronary syndromes; patients with higher levels of troponin on admission or a peak of troponin measured within 48 hours of a myocardial infarction are likely to have a more difficult and prolonged recovery than patients with lower troponin levels (Wettersten & Maisel, 2015).

Prior studies have shown a circadian rhythm influence on cytokine expression (Beglinger et al., 2017; Scheiermann et al., 2013). Because 43% of the identified biomarkers in this review are related to immune response; future studies should control the time of day the blood is collected for biomarker determination. Finally, the time of the blood sample collection was constrained to within 24±6 hrs of stroke onset in this review. These early markers may be used in a predictive way to identify subjects at high risk for poor recovery beyond the discharge NIHSS and mRS, so they could be targeted for more aggressive interventions, Additionally, future investigations may extend the time of blood collection to 3 days, 7 days, or even 3 months after stroke to help understand how molecules released in the later response phase correlate with physical outcome after ischemic stroke.

**Summary/Conclusions**

A systematic review of biomarkers for prediction of stroke recovery has not been updated since 2011. In the present review, 34 blood biomarkers were significantly associated with physical outcome after ischemic stroke. These biomarkers fall into the biological classifications of immune response, lipid/metabolism, neuronal function, and
blood vessel/circulation. The majority of biomarkers appears to predict physical recovery at 3 months following ischemic stroke, with fewer biomarkers predictive of recovery at 1 and 6 months.

**Author Contributions**

All authors contributed to the design of the study, development of the search strategy, establishment of the inclusion and exclusion criteria, data extraction criteria, analyses, and interpretation. YJL performed the study search, screening, and extraction of data. In addition, YJL wrote the first draft of the manuscript, and SKH, RC, JW, and LDM provided critical revision of the paper. All authors read and approved the final manuscript.

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References


*Circulation, 137*(12), e67-e492. doi:10.1161/CIR.0000000000000558

doi:10.1016/j.clim.2011.02.001


doi:10.1042/cs20100427


doi:10.1007/s11606-011-1966-4

from the CoRisk study. *Neurology, 80*(14), 1278-1286.

doi:10.1212/WNL.0b013e3182887944


doi:10.1212/WNL.0000000000004922


doi:10.1586/14737175.2015.1035712


doi:10.1161/CIRCRESAHA.110.223545


doi:10.1007/s00415-011-6379-0


in patients following ischemic stroke. *Translational Stroke Research, 8*(6), 578-584. doi:10.1007/s12975-017-0545-3

Saver, J. L., & Altman, H. (2012). Relationship between neurologic deficit severity and final functional outcome shifts and strengthens during first hours after onset. *Stroke, 43*(6), 1537-1541. doi:10.1161/STROKEAHA.111.636928


Table 1

**Categories of Biomarkers Significantly Associated with Physical Outcome after Ischemic Stroke**

<table>
<thead>
<tr>
<th>Category</th>
<th>Biomarker</th>
<th>Article Reference Numbers</th>
</tr>
</thead>
<tbody>
<tr>
<td>Immune response</td>
<td>Adiponectin, CCL11, Copeptin, CRP, GDF-15, IL-1β, IL-1Ra, IL-10, IL-12, IL-6, IL-8, MCP1, Osteopontin, TNFα, WBC</td>
<td>(Brea et al., 2011; De Marchis et al., 2013; De Marchis et al., 2018; Gori et al., 2017; Groschel et al., 2012; Kuwashiro et al., 2014; Mendioroz et al., 2011; Potpara et al., 2014; Rodriguez et al., 2013; Roy-O’Reilly et al., 2017)</td>
</tr>
<tr>
<td>Lipids/Metabolism</td>
<td>Cholesterol, Glucose, HDL-C, LDL-C</td>
<td>(Brea et al., 2011; Campos et al., 2011; De Marchis et al., 2013; De Marchis et al., 2018; Luitse et al., 2013; Makihara et al., 2012; Potpara et al., 2014)</td>
</tr>
<tr>
<td>Neuronal function</td>
<td>BDNF, Glutamate, GOT, GPT</td>
<td>(Campos et al., 2011; Lasek-Bal et al., 2015)</td>
</tr>
<tr>
<td>Blood vessel/Circulation</td>
<td>CrCl, D-Dimer, eGFR, Endostatin, Fibrinogen, Fibronectin, Hematocrit, Hemoglobin, MR-proANP, proMMP-10, Troponin</td>
<td>(Campos et al., 2011; De Marchis et al., 2013; De Marchis et al., 2018; Navarro-Sobrino et al., 2011; Potpara et al., 2014; Rodriguez et al., 2013)</td>
</tr>
</tbody>
</table>

*Note.* BDNF=Brain-derived neurotrophic factor; CCL11=C-C motif chemokine 11; CrCl=Creatinine Clearance; CRP=C-reactive protein; eGFR=Estimated glomerular filtration rate; GDF-15=Growth Differentiation Factor-15; GOT=Glutamic oxaloacetic transaminase; GPT=Glutamic pyruvic transaminase; HDL-C=High-density lipoprotein cholesterol; IL=Interleukin; LDL-C=Low-density lipoprotein cholesterol; MCP1=Monocyte chemoattractant protein 1; MMP=Matrix Metalloproteinase; MR-proANP=Midregional proatrial natriuretic peptide; TNFα=Tumor necrosis factor-alpha; WBC=White blood cell.
Table 2

*Biomarkers Significantly Associated with Physical Outcome after Ischemic Stroke by Time after Stroke.*

<table>
<thead>
<tr>
<th>Time after stroke</th>
<th>Biomarker associated with physical outcome</th>
<th>Article Reference Numbers</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 month</td>
<td>Immune response: CRP, WBC</td>
<td>Lipids/Metabolism: Cholesterol, LDL-C</td>
</tr>
<tr>
<td></td>
<td>Lipids/Metabolism: Glucose, HDL-C</td>
<td>Neuronal function: BDNF, Glutamate, GOT, GPT</td>
</tr>
<tr>
<td>3 months</td>
<td>Immune response: Adiponectin, CCL11, Copeptin, CRP, GDF-15, IL-1β, IL-1Ra, IL-6, IL-8, IL-10, IL-12, MCP1, Osteopontin, TNFα</td>
<td>Lipids/Metabolism: Glucose, HDL-C</td>
</tr>
<tr>
<td>6 months</td>
<td>CCL11</td>
<td>Glucose</td>
</tr>
<tr>
<td>----------</td>
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</tr>
</tbody>
</table>

*(Luitse et al., 2013; Roy-O’Reilly et al., 2017)*

**Note.** BDNF=Brain-derived neurotrophic factor; CCL11=C-C motif chemokine 11; CrCl=Creatinine Clearance; CRP=C-reactive protein; eGFR=Estimated glomerular filtration rate; GDF-15=Growth Differentiation Factor-15; GOT=Glutamic oxaloacetic transaminase; GPT=Glutamic pyruvic transaminase; HDL-C=High-density lipoprotein cholesterol; IL=Interleukin; LDL-C=Low-density lipoprotein cholesterol; MCP1=Monocyte chemoattractant protein 1; MMP=Matrix Metalloproteinase; MR-proANP=Midregional proatrial natriuretic peptide; TNFα=Tumor necrosis factor-alpha; WBC=White blood cell.
Figure 1. Flow diagram of the literature search. Adapted from “The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate healthcare interventions: explanation and elaboration” by Liberati, A. and Altman, D. G., 2009, British Medical Journal, 339, b2700.
Figure 2. Study quality of the 18 articles in the systematic review assessed by the modified REMARK questionnaire (Supplementary Table 4).
## Supplementary Material

### Supplementary Table 1

**Search Strategies in PubMed, Embase, and CINAH**

<table>
<thead>
<tr>
<th>Search strategies</th>
<th>PubMed</th>
</tr>
</thead>
</table>
(3) physical recovery


AND


(4) adult


These four search strategies were combined as follow: (1) and (2) and (3) and (4).

<table>
<thead>
<tr>
<th>Search strategies</th>
<th>Embase</th>
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<tbody>
<tr>
<td>(1) biomarker</td>
<td>immune markers:ti,ab,kw OR 'immune marker':ti,ab,kw OR 'immunologic markers':ti,ab,kw OR 'immunologic marker':ti,ab,kw OR 'blood biomarker':ti,ab,kw OR 'blood biomarkers':ti,ab,kw OR 'serum biomarker':ti,ab,kw OR 'serum biomarkers':ti,ab,kw OR 'serum factors':ti,ab,kw OR 'biologic marker':ti,ab,kw OR 'biologic markers':ti,ab,kw OR 'biomarker':ti,ab,kw OR 'biomarkers':ti,ab,kw OR 'biologic marker':ti,ab,kw OR 'biomarker':ti,ab,kw OR 'biologic markers':ti,ab,kw OR 'biomarkers':ti,ab,kw OR 'biologic marker':ti,ab,kw OR 'biomarker':ti,ab,kw</td>
</tr>
</tbody>
</table>
(2) ischemic stroke    cerebrovascular disease'/exp OR 'cerebrovascular disorders':ti,ab,kw OR 'cerebrovascular disorder':ti,ab,kw OR 'cerebrovascular':ti,ab,kw OR 'brain ischemia'/exp OR 'brain ischemia':ti,ab,kw OR 'brain ischamias':ti,ab,kw OR 'ischemic stroke':ti,ab,kw OR 'ischemic strokes':ti,ab,kw OR 'cva':ti,ab,kw OR 'stroke syndrome':ti,ab,kw OR 'stroke syndromes':ti,ab,kw OR 'cerebral vascular accident':ti,ab,kw OR 'cerebral vascular accidents':ti,ab,kw OR 'cerebrovascular accident':ti,ab,kw OR 'cerebrovascular accidents':ti,ab,kw OR 'brain ischamias':ti,ab,kw OR 'cerebral ischemia':ti,ab,kw OR 'cerebral ischamias':ti,ab,kw OR 'ischemic brain':ti,ab,kw OR 'ischemic brains':ti,ab,kw

(3) physical recovery    functional recovery'/exp OR 'prognosis'/exp OR 'stroke rehabilitation'/exp OR 'functional recovery':ti,ab,kw OR 'prognosis':ti,ab,kw OR 'stroke rehabilitation':ti,ab,kw OR 'recovery of function':ti,ab,kw OR 'function recovery':ti,ab,kw OR 'function recoveries':ti,ab,kw OR 'physical recovery':ti,ab,kw OR 'physical recoveries':ti,ab,kw OR 'functional recoveries':ti,ab,kw OR 'stroke recovery':ti,ab,kw OR 'stroke recoveries':ti,ab,kw OR 'motor recovery':ti,ab,kw OR 'motor recoveries':ti,ab,kw OR 'prognoses':ti,ab,kw OR 'stroke rehabilitations':ti,ab,kw

AND
'tfunctional independence':ti,ab,kw OR 'disability':ti,ab,kw OR 'severity':ti,ab,kw OR 'mobility':ti,ab,kw OR 'physical independence':ti,ab,kw OR 'motor functions':ti,ab,kw OR 'motor function':ti,ab,kw OR 'health outcomes':ti,ab,kw OR 'health outcomes':ti,ab,kw OR 'physical outcomes':ti,ab,kw OR 'functional outcomes':ti,ab,kw OR 'motor performance':ti,ab,kw OR 'motor performances':ti,ab,kw OR 'physical function':ti,ab,kw OR 'physical functions':ti,ab,kw

(4) adult    adult'/exp OR 'aged'/exp OR 'adult':ti,ab,kw OR 'adults':ti,ab,kw OR 'aged':ti,ab,kw OR 'elderly':ti,ab,kw OR 'aged 18 years':ti,ab,kw OR 'age 18 years':ti,ab,kw OR 'aged 65 years':ti,ab,kw OR 'age 65 years':ti,ab,kw OR 'working age':ti,ab,kw OR 'working aged':ti,ab,kw

These four search strategies were combined as follow: (1) and (2) and (3) and (4).

<table>
<thead>
<tr>
<th>CINAHL</th>
<th>Search strategies</th>
<th>Search keywords</th>
</tr>
</thead>
<tbody>
<tr>
<td>(1) biomarker</td>
<td>(MH &quot;Biological Markers&quot;) OR &quot;Biomarkers&quot; OR &quot;Biomarker&quot; OR &quot;Immune Markers&quot; OR &quot;Immune Marker&quot; OR &quot;immunologic Markers&quot; OR &quot;immunologic Marker&quot; OR &quot;blood biomarker&quot; OR &quot;blood biomarkers&quot; OR &quot;serum biomarker&quot; OR &quot;serum biomarkers&quot; OR &quot;plasma biomarker&quot; OR &quot;plasma biomarkers&quot; OR &quot;plasma marker&quot; OR &quot;plasma markers&quot; OR &quot;serum marker&quot; OR &quot;serum markers&quot; OR &quot;blood marker&quot; OR &quot;blood markers&quot; OR &quot;blood indicator&quot; OR &quot;blood indicators&quot; OR &quot;serum indicator&quot; OR &quot;serum indicators&quot; OR &quot;plasma indicator&quot; OR &quot;plasma indicators&quot; OR &quot;plasma factors&quot; OR &quot;plasma factor&quot; OR &quot;serum factor&quot; OR &quot;serum factors&quot; OR &quot;blood factors&quot; OR &quot;blood factor&quot; OR &quot;biological marker&quot; OR &quot;biological markers&quot; OR &quot;biologic markers&quot; OR &quot;biologic marker&quot; OR (MH &quot;Cytokines&quot;) OR (MH &quot;Chemokines&quot;) OR &quot;cytokine&quot; OR &quot;cytokines&quot; OR &quot;chemokines&quot; OR &quot;chemokine&quot;</td>
<td></td>
</tr>
</tbody>
</table>
(2) ischemic stroke MH "Cerebrovascular Disorders" OR "Cerebrovascular Disorders" OR "Cerebrovascular Disorder" OR "Cerebrovascular" OR "ischemic stroke" OR "Brain Ischemia" OR "cva" OR "ischemic strokes" OR "stroke syndrome" OR "cerebral vascular accident" OR "cerebrovascular accident" OR "cerebrovascular accidents" OR "stroke syndromes" OR "cerebral vascular accidents" OR (MH "Cerebral Ischemia") OR "Brain Ischemias" OR "cerebral ischemia" OR "cerebral ischamias" OR "ischemic brain" OR "ischemic brains"

(3) physical recovery "Recovery of Function" OR "function recovery" OR "function recoveries" OR "physical recovery" OR "physical recoveries" OR "functional recovery" OR "functional recoveries" OR "stroke recovery" OR "motor recovery" OR "motor recoveries" OR MH "Prognosis" OR "Prognosis" OR "Prognoses" OR "Stroke Rehabilitation" OR "Stroke Rehabilitations" AND "severity" OR "disability" OR "mobility" OR "functional independence" OR "physical independence" OR "motor function" OR "motor functions" OR "health outcome" OR "health outcomes" OR "physical outcome" OR "physical outcomes" OR “functional outcomes” OR “functional outcome” OR “motor performance” OR “motor performances” OR “physical function” OR “physical functions”

(4) adult (MH "Adult") OR "Adult" OR "adults" OR (MH "Aged") OR "Aged" OR "aged 18 years" OR "age 18 years" OR "elderly" OR "aged 65 years" OR "age 65 years" OR "working age" OR "working-age" OR "working aged" OR "working-aged"

These four search strategies were combined as follow: (1) and (2) and (3) and (4).
### Supplementary Table 2

**REMARK Quality Questionnaire**

<table>
<thead>
<tr>
<th>Item</th>
<th>Yes</th>
<th>No</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>1 Was the study prospective?</strong></td>
<td></td>
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<tr>
<td>YES: The study reported that patients and blood samples were collected prior to the development of an outcome</td>
<td></td>
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<tr>
<td>NO: No report or clearly retrospective (e.g. patients with poor prognosis collected prior to biomarker measurement)</td>
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<tr>
<td><strong>2 Was the evaluation of prognostic marker blinded to patient outcome?</strong></td>
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<tr>
<td>YES: The study reported an attempt to blind the person measuring the level of biomarker to patient outcome</td>
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<tr>
<td>NO: There was no such report</td>
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<tr>
<td><strong>3 Was there a defined time period during which patients were enrolled?</strong></td>
<td></td>
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<tr>
<td>YES: Study defined time period, end of follow up period and median follow up time</td>
<td></td>
<td></td>
</tr>
<tr>
<td>NO: Did not define above criteria</td>
<td></td>
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<tr>
<td><strong>4 Were there precisely defined clinical outcomes at the beginning of the study?</strong></td>
<td></td>
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<tr>
<td>YES: Study defined which clinical endpoints are to be measured</td>
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<tr>
<td>NO: No such definition</td>
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<tr>
<td><strong>5 Did the study provide a rationale for study sample size?</strong></td>
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<tr>
<td>YES: Evidence of a sensible sample size calculation</td>
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<tr>
<td>NO: No attempt to define sample size</td>
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<tr>
<td><strong>6 Did the study provide a list of candidate variables?</strong></td>
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<tr>
<td>YES: A list of variables to be considered in multiple regression analysis were provided at the beginning of the study</td>
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<tr>
<td>NO: Evidence that variables measured and not reported</td>
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<tr>
<td><strong>7 Were the methods for measuring the prognostic marker adequately described and referenced?</strong></td>
<td></td>
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<tr>
<td>YES</td>
<td></td>
<td></td>
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<tr>
<td>NO</td>
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<tr>
<td><strong>8 Were the characteristics of the study patients described?</strong></td>
<td></td>
<td></td>
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<tr>
<td>YES: The study described the source and inclusion and exclusion criteria.</td>
<td></td>
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</tr>
</tbody>
</table>
| NO: Did not provide the information or it was unclear description.
Note. REMARK= REporting recommendations for tumour MARKer prognostic studies. Modified from “Blood markers for the prognosis of ischemic stroke: A systematic review” by Whiteley et al., 2009, Stroke, 40(5), e380-389(Whiteley et al., 2009).
**Supplementary Table 3**

*Characteristics of Studies that Examined Blood Biomarkers on Physical Recovery in Ischemic Stroke*

<table>
<thead>
<tr>
<th>Author, year</th>
<th>Sample collection Country, year</th>
<th>Cohort</th>
<th>Patient number (male %)</th>
<th>Patient age (mean ± SD)</th>
<th>Blood biomarkers</th>
<th>Outcome measures (time point)</th>
<th>Results: blood biomarker levels according to functional outcomes.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Navarro-Sobrino et al., 2011 (Navarro-Sobrino et al., 2011)</td>
<td>Spain</td>
<td>Healthy controls vs Ischemic stroke (with tPA treatment)</td>
<td>26 (46.2%) vs 109 (55%)</td>
<td>69±9.5 vs 70.9±15.1</td>
<td>*Endostatin SDF-1 HCG KGF HGF TSP-1 TSG-1 VEGF VEGF-R2 PDGF-BB PDGF-AA Angiostatin</td>
<td>mRS (3M) NIHSS (Admission, 1h, 2h, 12h, 24h, 48h and discharge)</td>
<td>1. Higher endostatin is associated with poor outcome (mRS≥3). 2. Lower KGF/endostatin, KGF/TSP-1, and VEGF-R2/TSP-1 are associated with worse severity (admission). 3. Lower KGF/TSP-1 and VEGF-R2/TSP-1 are associated with worse severity (1h). 4. Lower HCG, HCG/endostatin, KGF/endostatin, KGF/TSP-1, VEGF, PDGF-BB/Angiostatin, PDGF-AA/TSG-1, VEGF/TSP-1, VEGF/endostatin, VEGF-R2/TSP-1, and HGF/Angiostatin are associated with worse severity (2 and 12h). 5. VEGF, PDGF-BB/Angiostatin, PDGF-AA/TSG-1, VEGF/endostatin, and HGF/Angiostatin are associated with worse severity (24h). 6. VEGF, PDGF-BB/Angiostatin, and VEGF/endostatin are associated with worse severity (48h and discharge).</td>
</tr>
<tr>
<td>Study</td>
<td>Country, Period</td>
<td>Study Type</td>
<td>Control Group</td>
<td>Case Group</td>
<td>Measures</td>
<td>mRS</td>
<td>NIHSS (Admission)</td>
</tr>
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</tr>
<tr>
<td>Mendioroz et al., 2011</td>
<td>Spain, Dec. 2006 - June 2008</td>
<td>Healthy controls vs Ischemic stroke (with tPA treatment)</td>
<td>40 (46.2%) vs 178 (52.8%)</td>
<td>71.5±13.4 vs 71.91±8.1</td>
<td>*Osteopontin Glucose mRS (3M) NIHSS (Admission)</td>
<td></td>
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</tr>
<tr>
<td>Brea et al., 2011</td>
<td>Spain, Feb. 2009 - Aug 2009</td>
<td>Ischemic stroke</td>
<td>110 (58.2%) mRS&lt;3: 48 mRS≥3: 62</td>
<td>74.6±9.8 mRS&lt;3: 71.9±8.9 mRS≥3: 72.9±10.9</td>
<td>*Glucose WBC Platelets Fibrinogen *CRP TLR3 TLR7 TLR8 TLR9 mRS (3M) NIHSS (Admission, 24h, 72h, 7 days, 3M)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Campos et al., 2011</td>
<td>Spain</td>
<td>First episode of ischemic stroke</td>
<td>365 (57.5%) mRS&lt;3: 168 mRS≥3: 197</td>
<td>70.5±11.4</td>
<td>*Glucose WBC Platelets *Fibrinogen *Glutamate *GOT *GPT mRS (3M) NIHSS (Admission)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Groschel et al., 2012</td>
<td>Germany, Mar. 2009 - Feb. 2010</td>
<td>Ischemic stroke</td>
<td>264 (55.3%)</td>
<td>70.3±12.7</td>
<td>*GDF-15 WBC CRP Cholesterol LDL-C HDL-C Triglyceride mRS (3M) NIHSS (Admission)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1. Higher osteopontin is associated with poor outcome (mRS≥3).
2. No relationship between glucose and stroke disability (mRS≥3).
3. No relationship between osteopontin and stroke severity.

1. Higher glucose and CRP are associated with poor outcome (mRS≥3).
2. TLR3, 7, 8, and 9 are not associated with stroke disability.

1. Higher glucose, fibrinogen, and blood glutamate are associated with poor outcome (mRS≥3).
2. Lower GOT and GPT are associated with poor outcome (mRS≥3).

1. Higher GDF-15 is associated with poor outcome (mRS≥2).
2. Higher GDF-15 is associated with worse severity.
<table>
<thead>
<tr>
<th>Study</th>
<th>Location</th>
<th>Type of Stroke</th>
<th>Treatment</th>
<th>mRS &lt;2</th>
<th>mRS ≥2</th>
<th>Total Cholesterol</th>
<th>mRS (3M)</th>
<th>NIHSS (Admission)</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Makihara et al., 2012 (Makihara et al., 2012)</td>
<td>Japan, Oct. 2005-July 2008</td>
<td>Ischemic stroke with tPA treatment</td>
<td>489 (65%)</td>
<td>188</td>
<td>301</td>
<td>HDL-C</td>
<td>70.8±11.6</td>
<td></td>
<td>1. Lower HDL-C is associated with poor outcome (mRS≥2).</td>
</tr>
<tr>
<td>Delgado et al., 2012 (Delgado et al., 2012)</td>
<td>Spain</td>
<td>Healthy controls vs Ischemic stroke</td>
<td>135</td>
<td>99 (51%)</td>
<td>72</td>
<td>Lp-PLA2 mass</td>
<td>mRS (3M)</td>
<td>NIHSS (Admission)</td>
<td>1. Neither Lp-PLA2 mass or Lp-PLA2 activity is not associated with stroke severity. 2. No differences were found in either Lp-PLA2 mass or activity according to the third month.</td>
</tr>
<tr>
<td>Rodriguez et al., 2013 (Rodriguez et al., 2013)</td>
<td>Spain</td>
<td>Healthy controls vs Ischemic stroke</td>
<td>With tPA: 76 (58%)</td>
<td>With tPA: 66.9±11.3</td>
<td>Without tPA: 73.5±11.3</td>
<td>MMP9 *C-Fibronectin *IL-6 *TNFα</td>
<td>mRS (3M)</td>
<td>NIHSS (Admission, 24h, 48h)</td>
<td>1. proMMP-10, C-Fibronectin, IL-6, and TNFα are significantly higher in tPA treated patients with poor outcome (mRS≥3). *proMMP-10 is significantly higher in non-tPA treated patients with poor outcome (mRS≥3).</td>
</tr>
<tr>
<td>Luitse et al., 2013 (Luitse et al., 2013)</td>
<td>Netherlands, Jan. 2007-June 2008</td>
<td>Ischemic stroke (NG vs HG)</td>
<td>Total: 80</td>
<td>NG: 47 (58.8%)</td>
<td>HG: 33 (41.3%)</td>
<td>*Glucose</td>
<td>mRS (6M)</td>
<td>NIHSS (Admission)</td>
<td>1. Hyperglycemia is associated with poor outcome (mRS≥3). 2. Hyperglycemia is associated with worse severity (no statistical data).</td>
</tr>
<tr>
<td>Study</td>
<td>Country</td>
<td>Duration</td>
<td>Diagnosis</td>
<td>Total: 783</td>
<td>Without tPA:</td>
<td>With tPA:</td>
<td>mRS (3M)</td>
<td>NIHSS (Admission)</td>
<td>Adverse Outcomes</td>
</tr>
<tr>
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<tr>
<td>De Marchis et al., 2013 (De Marchis et al., 2013)</td>
<td>Germany and Switzerland, Mar. 2009-Apr. 2011</td>
<td>Ischemic stroke</td>
<td>71.0 (60.6-80.0)</td>
<td>465 (59.4%)</td>
<td>318 (40.6%)</td>
<td></td>
<td></td>
<td></td>
<td>1. Higher copeptin, glucose, and CRP are associated with poor outcome (mRS≥3).</td>
</tr>
<tr>
<td>Selçuk et al., 2014 (Selçuk et al., 2014)</td>
<td>Turkey, May 2011-Oct. 2011</td>
<td>Ischemic stroke</td>
<td>68±13</td>
<td>50 (48%)</td>
<td>S100B</td>
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<td>1. The first day S100B level is not associated with post-stroke disability at 1 month.</td>
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<td>2. There was a poor correlation between functional outcome at 1-month post-stroke and the third day S100B level.</td>
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<td></td>
<td>3. No correlation between stroke severity and S100B level.</td>
</tr>
<tr>
<td>Potpara et al., 2014 (Potpara et al., 2014)</td>
<td>UK</td>
<td>Ischemic stroke</td>
<td>70.0±8.9</td>
<td>240 (57.9%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1. Higher CRP, fibrinogen, cardiac Tnl, D-dimer, WBC, LDL, and total cholesterol are associated with poor outcome (mRS≥3).</td>
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<td>2. Lower CrCl and hematocrit are associated with poor outcome.</td>
</tr>
<tr>
<td>Study</td>
<td>Location</td>
<td>Population Description</td>
<td>Sample Size</td>
<td>mRS (3M)</td>
<td>NIHSS (Admission)</td>
<td></td>
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<tr>
<td>Kuwashiro et al., 2014(Kuwashiro et al., 2014)</td>
<td>Japan, Nov. 2007-Apr. 2010</td>
<td>Healthy controls vs Ischemic stroke</td>
<td>342 (67.3%)</td>
<td>68.3±10.1</td>
<td>*Adiponectin</td>
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<tr>
<td>Bustamante et al., 2014(Bustamante et al., 2014)</td>
<td>Spain, Mar. 2003-Nov 2005</td>
<td>Ischemic stroke (with tPA treatment)</td>
<td>159 (55.3%)</td>
<td>70.1±11.4</td>
<td>ChT</td>
<td></td>
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<tr>
<td>Lasek-Bal et al., 2015(Lasek-Bal et al., 2015)</td>
<td>Poland, June 2014-April 2015</td>
<td>First episode of ischemic stroke (with tPA treatment)</td>
<td>87 (51.7%)</td>
<td>71.7±11.8</td>
<td>*BDNF</td>
<td></td>
<td></td>
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<tr>
<td>Roy-O’Reilly et al., 2017(Roy-O’Reilly et al., 2017)</td>
<td>USA, 2011-2015</td>
<td>Ischemic stroke</td>
<td>133 (57.1%)</td>
<td>70.42±13.87</td>
<td>*CCL11</td>
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</tbody>
</table>

1. Higher adiponectin is associated with worse severity and poor outcome (mRS≥3).

2. ChT activity is not related to baseline stroke severity.

2. Higher ChT activity is associated with poor outcome (mRS≥3), but ChT activity is not an independent predictor.

1. Lower BDNF is associated with poor outcome at 3 months post-stroke (mRS≥3).

1. Lower CCL11 is associated with poor outcome at 3 months (mBI≤14) and 12 months (mRS≥3) post-stroke.
<table>
<thead>
<tr>
<th>Study</th>
<th>Location</th>
<th>Duration</th>
<th>Type of Stroke</th>
<th>Reference</th>
<th>n</th>
<th>Mean mRS</th>
<th>NIHSS (Admission)</th>
<th>Additional Information</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gori et al., 2017 (Gori et al., 2017)</td>
<td>Italy, Oct. 2008-June 2011</td>
<td>Ischemic stroke (with tPA treatment)</td>
<td>327 (58.1%)</td>
<td>68.9±12.1</td>
<td>mRS (3M)</td>
<td>NIHSS (Admission)</td>
<td>*IL-1β, *IL-1RA, IL-4, *IL-6, *IL-8, *IL-10, *IL-12, IL-17, IFNγ, IP10, *MCP1, MIP1β, *TNFα, *A2M, SAP, Haptoglobin, MMP1, 2, 3, 7, 8, and 9, TIMP1, 2, and 4</td>
<td>1. CRP, IL-1β, IL-1Ra, IL-6, IL-8, IL-10, IL-12, TNFα, and MCP1 (Pre-post tPA) were associated with three-month (mRS≥3).</td>
</tr>
</tbody>
</table>

* indicates statistically significant difference; A2M=Alpha-2-Macroglobulin; BDNF=Brain-derived neurotrophic factor; CCL11=C-C motif chemokine 11; ChT=Chitotriosidase; CrCl = Creatinine Clearance; CRP = C-reactive protein; eGFR = Estimated glomerular filtration rate; GDF-15 = Growth Differentiation Factor-15; GOT=Glutamic oxaloacetic transaminase; GPT=Glutamic pyruvic transaminase; h=hour(s); HDL-C=High-density lipoproteins cholesterol; HG=Hyperglycaemia; HGF=Hepatocyte growth factor; IFNγ=Interferon gamma; IL=Interleukin; IP10=Interferon gamma-induced protein 10; KGF=Keratinocyte growth factor; LDL-C=Low-density lipoproteins cholesterol; Lp-PLA2=Lipoprotein-associated phospholipase A2; M=month(s); mBI= modified Barthel index; MCP1=Monocyte chemoattractant protein 1; MIP1β=Macrophage inflammatory protein-1β; MMP=Matrix Metalloproteinase; MR-proANP=Midregional proatrial natriuretic peptide; mRS=modified Rankin score; NG=Normoglycaemia; NIHSS=National Institute of Health Stroke Scale.
Institutes of Health Stroke Scale; PDGF=Derived growth factor; S100B=S100 calcium-binding protein B; SAP=Serum amyloid P-component; SD=Standard deviation; SDF-1=Stromal cell-derived factor-1; TIMP-1=Tissue inhibitor of matrix metalloproteinases-1; TLR=Toll-like receptor; TNFα=Tumor necrosis factor-alpha; TnI=Troponin I; tPA=Tissue plasminogen activator; TSG-1=Tumor necrosis factor-inducible gene-1; TSP-1=Thrombospondin-1; VEGF=Vascular endothelial growth factor; W=week(s); WBC=White blood cell.
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*Note.* REMARK=REporting recommendations for tumour MARKer prognostic studies.
Supplemental References

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Relationships among Optimism, Inflammation, and Stroke Recovery

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Abstract

**Objective:** Post-stroke inflammation is detrimental to the brain and results in an unfavorable recovery. Optimism has been associated with lower inflammation and better health outcomes among people with medical conditions, but no studies have assessed this association in the stroke population. The overall goals were to examine the relationships among optimism, stroke severity, physical disability, and inflammation during hospitalization and evaluate the relationships among optimism, inflammation, and stroke recovery over the three-month post-stroke period.

**Methods:** This study was a secondary analysis of data prospectively collected from the BioRepository of Neurological Diseases biobank. Outcomes included optimism, stroke severity, physical recovery, and inflammatory markers (IL-6, TNF-α, and CRP). Spearman’s correlation, Wilcoxon signed-rank test, multiple linear regression, and mixed-effect regression model were used to determine the relationships among the variables.

**Results:** A total of 49 subjects at baseline, with 13 at 3-month follow-up were recruited. The results indicated that subjects with higher optimism showed less stroke impairment and lower level of CRP at baseline compared to those with lower optimism. Additionally, optimism was associated with less stroke severity and lower IL-6 and CRP levels over the first three months after stroke.

**Conclusion:** Optimistic stroke survivors showed lower inflammation and better stroke recovery. By understanding this relationship may provide a scientific framework whereby new strategies for stroke recovery can be developed in the future.

**Keywords:** optimism, inflammation, stroke recovery
Acronyms: LOT-R = revised Life Orientation Test; NIHSS = National Institutes of Health Stroke Scale; mRS = modified Rankin Scale; IL-6 = Interleukin 6; TNF-α = Tumor necrosis factor alpha; CRP = C-Reactive Protein.
Introduction

Stroke is the 5th leading cause of death in the U.S., with an annual incidence of approximately 795,000 cases (Benjamin et al., 2019). Ischemic stroke accounts for 87% of all strokes. The most common neurological disease in the adult population worldwide, stroke is now the primary cause of long-term adult disability (Feigin et al., 2017); up to 74% of stroke survivors are dependent in activities of daily living (Miller et al., 2010). Motor impairment, sensory dysfunction, and dysphasia are common manifestations of stroke. Approximately 30% of stroke survivors in the U.S. suffer permanent disability, and 20% of survivors require inpatient rehabilitation within three months after stroke (Creutzfeldt et al., 2012).

Improvements in acute stroke care have reduced stroke-related mortality over the past two decades. However, the increased survival rate leaves many survivors living with severe disability, placing a tremendous burden on their caregivers and the healthcare system (F. Mu et al., 2017). Post-stroke rehabilitation is a continuum, starting within days of stroke onset and ending only when it no longer produces any positive effect. Physical recovery includes motor function, sensation, language and swallowing ability (Harvey, 2015). Prior studies indicated that 48 - 91% of physical recovery occurred within the first three months after stroke (Lee et al., 2015; Stinear, 2017; Wade et al., 1985).

Ischemic stroke caused by blood vessel blockage results in cell damage and/or cell death. These damaged cells may evoke a massive upregulation of pro-inflammatory cytokines, such as interleukin 6 (IL-6), tumor necrosis factor alpha (TNF-α), and C-reactive protein (CRP), which attract infiltration of innate immune cells from the circulatory system to site of the damaged tissue and support post-stroke healing (Dirnagl
et al., 1999; Lakhan et al., 2009; Simats et al., 2016). Even though the initial immune response is to facilitate repair and regeneration of the damaged tissue, the infiltrated immune cells may produce cytotoxic mediators that prolong the inflammatory response, exacerbate brain damage, and contribute to edema formation and hemorrhagic transformation (Hu et al., 2016). These secondary complications commonly influence stroke outcomes and leave an individual with residual impairment of physical, psychological, and cognitive functions. Hence, properly regulating post-stroke inflammation may be a therapeutic strategy to reduce secondary brain injury and enhance post-stroke recovery (Simats et al., 2016).

Optimism is conceptually defined as positive expectations of the future (Scheier & Carver, 1985). It is considered a personality trait and a positive psychological factor (Conversano et al., 2010a; Scheier & Carver, 1985). Studies have shown that optimism may change over time or during situational changes, such as in times of high stress (Atienza et al., 2004; Segerstrom, 2007). A meta-analysis indicated that psychological intervention approaches such as Best Possible Self (BPS) or cognitive-behavior therapy (CBT) might significantly augment optimism levels among various populations (Malouff & Schutte, 2017).

Accumulated studies have found that optimism improves both mental and physical health outcomes. For example, optimistic people have reported greater resilience to stress, better recovery after cardiac bypass surgery, and improved functional outcomes after traumatic brain injury (Ramanathan et al., 2011; Ronaldson et al., 2014; Tindle et al., 2012). Additionally, inflammation is prevalent after critical illness and is associated with poor physical recovery during the first three months of post-intensive care unit
discharge (Griffith et al., 2016). Optimism has been linked to lower levels of chronic inflammation in older adults (Ikeda et al., 2011) and less blood pro-inflammatory cytokines, such as IL-6, CRP, and fibrinogen, in patients with cardiovascular disease (Roy et al., 2010).

A qualitative study found that optimistic stroke survivors may exhibit more confidence to face the challenges of stroke recovery (Jones et al., 2008). Moreover, a recent study, in which the online survey was conducted by 176 stroke survivors, also showed that up to 41 years since diagnosis of stroke, the survivors with higher levels of optimism reported fewer depressive symptoms and better perceived overall health compared to those with lower levels of optimism (Shifren & Anzaldi, 2018b). However, no studies have examined the links among optimism, inflammation, and physical recovery in the stroke population.

The purpose of this study is to examine the relationship among optimism, inflammation, and physical recovery in the stroke population. Given the evidence of associations among optimism, inflammation, and physical health in multiple other diseases, but not stroke, determining how optimism associated with inflammation and post-stroke recovery is important. Understanding the mechanisms underlying the role of optimism in post-stroke inflammation and physical recovery could allow for the development of psychological interventions for boosting optimism. The specific aims and hypotheses of this study were: (1) Examine the relationships among optimism, stroke severity, physical disability, and inflammation during hospitalization. It was hypothesized that stroke patients with higher levels of optimism during hospitalization would have lower levels of stroke severity, physical disability, and pro-inflammatory cytokines
compared to those with lower optimism levels. (2) Examine the longitudinal relationships among optimism, inflammation, and stroke recovery at three months post-stroke. It was hypothesized that patients’ levels of optimism would remain stable over the three months, and more optimistic patients will have less inflammation and accelerated stroke recovery over the 3-month post-stroke period compared with less optimistic patients.

Materials and Methods

Study Design

This pilot study was a secondary analysis of data prospectively collected from the BioRepository of Neurological Diseases (HSCMH-17-0452, Appendix A). A quantitative observational design with repeated measures was used.

Study Sample

The sample was selected from consecutive acute stroke subjects admitted to the Memorial Hermann Southwest Hospital from June 2018 to January 2019. To investigate the relationships among optimism, stroke severity, physical disability, and inflammation over a three-month post-stroke period, this study included subjects who were: (1) 18 years or older; (2) diagnosed with an ischemic stroke. Subjects were excluded if they (1) were diagnosed with transient ischemic attack or intracerebral hemorrhage, including secondary hemorrhage on control computed tomography or magnetic resonance imaging (to avoid confounding effects of hematoma-induced inflammation); (2) had underlying vascular lesions or traumatic brain injury, systemic malignancy, autoimmune disease, or immunosuppression use of any immunosuppressive drugs (defined as the use of immunosuppressive drugs for ≥28 days); and (3) could not provide informed consent.
Measurements

**Optimism.** The main predictor variable was the level of optimism, which was measured via the LOT-R (Appendix C) (Scheier et al., 1994). Scores on the LOT-R range from 0 to 24, with higher scores indicating higher levels of optimism. It includes 10 items, four of which are filler items that were not used in scoring. Of the six items that were scored, three are keyed in a positive direction (1, 4, and 10) and three in a negative direction (3, 7, and 9). The respondents indicate the extent to which they agree with each item on a 5-point Likert scale that ranges from strongly disagree to strongly agree.

Internal consistency of the LOT-R was reported $\alpha = 0.82$ (Shifren & Anzaldi, 2018b) in the stroke population and stability (test-retest reliability) over 4 months in college students was reported as $r = 0.79$ (Scheier et al., 1994). Cronbach’s alpha in this study was 0.72. Stability over the 3-month period was $r = 0.79$ ($p = 0.001$). In order to make appropriate comparisons and fully explore the effects of optimism and pessimism on stroke severity, physical disability, and inflammatory factors, three partly overlapping measures were developed: full scale with six items (LOT-R), optimism subscale with three items (OPT), and pessimism subscale with three items (PESS). The higher values indicate more positive orientation on LOT-R and OPT and more negative orientation on PESS.

**Clinical outcomes.** Stroke severity was evaluated via the NIHSS (Appendix D) by a certified neurologist (Brott et al., 1989). Scores on the NIHSS range from 0 to 42, with lower scores indicating less impairment. The acceptable interrater reliability was reported as an intraclass correlation coefficient of 0.82 (Goldstein & Samsa, 1997) and high content validity in the stroke population (Kasner, 2006b). Physical disability after
stroke was defined according to the mRS (Appendix E) by a certified neurologist (Banks & Marotta, 2007). Scores on the mRS range from 0 to 6, with lower scores indicating less disability. The acceptable internal consistency of mRS was reported as Cronbach’s α of 0.89 in stroke survivors (Wei et al., 2015).

Inflammatory markers. Peripheral blood samples were obtained from subjects at baseline and 3-month after stroke. Plasma was separated by centrifugation at 1,000-2,000xg for 10 minutes, aliquoted and stored at −80°C until used. Samples were coded and assayed in a blind manner regarding the subjects’ level of optimism, stroke severity (NIHSS), and physical disability (mRS). Levels of IL-6, TNF-α, and CRP were measured from plasma samples using enzyme-linked immunosorbent assay (ELISA) kits (D6050, HSTA00E, and DCRP00; R&D Systems, Minneapolis, MN). The assay sensitivity has been reported to be 0.7 pg/mL for IL-6, 0.049 pg/mL for TNF-α, and 0.022 ng/mL for CRP. Intra- and inter-assay percent coefficients of variation have been shown between 2.0% and 6.5%, indicating a high sensitivity and precision. In this study, the intra- and inter-assay percent coefficients of variation of IL-6, TNF-α, and CRP were less than 4.5%.

Data Collection

Data were obtained at baseline and 3-month follow-up visit, including LOT-R, NIHSS, mRS, IL-6, CRP, and TNF-α. At baseline, subjects’ demographic and clinical data, including age, gender, race/ethnicity, medical history, and social support (Need of Support and Service Questionnaire, NSSQ) were extracted from the BioRepository dataset. Researchers provided up to three telephone reminders in a week before and after the time of 3-month follow-up visit. All the data were de-identified before being
Identifiers were restricted to the medical record number, and this unique code was used to identify the patients in the data collection sheet based solely on their age, gender, and diagnosis of ischemic stroke. This study was approved by the UTHealth Committee for the Protection of Human Subjects (HSC-MS-18-0534, Appendix B).

**Sample Size Determination**

For Aim 1, we assumed that the minimum correlation coefficient between optimism and stroke severity, physical disability, and inflammation was 0.4 (Ikeda et al., 2011; Roy et al., 2010). To achieve a correlation coefficient with 80% power at type I error of 0.05, we would need 46 patients (Bujang & Baharum, 2016). For Aim 2, we planned to compare the outcomes of stroke recovery and inflammation between optimistic and non-optimistic groups. Based on related articles and our preliminary data, we assumed that the mean differences will be 8.2 with a standard deviation (SD) of 8.1 (effect size of 1.01) for scores on the NIHSS, 2.3 with a SD of 1.7 (effect size of 1.35) for scores on the mRS, and 1.8 with a SD of 2.2 (effect size of 0.82) for CRP levels. The required sample sizes for the three variables were 24, 14, and 36, respectively, to achieve 80% power at a type I error of 0.05 using longitudinal data analysis with a within-subject correlation of 0.5 (Hedeker et al., 1999).

**Data Analysis**

Statistical analyses were performed using SPSS for Windows, version 25.0 (SPSS Inc., Chicago, IL, USA). Descriptive statistics were used to analyze demographic characteristics and clinical history. Kolmogorov–Smirnov tests were performed to assess normality distributions in continuous variables. Correlations between numeric variables were assessed by Spearman correlation coefficient. Univariate regression analyses were
performed to model functional outcomes at baseline on variables (age, sex, race/ethnicity, and social support) one by one. Variables with a p-value less than 0.1 were then included in multiple linear models as confounders in evaluating the effect of optimism on baseline functional outcomes. To compare levels of optimism and other clinical outcomes between baseline and 3-month after stroke, Wilcoxon signed-rank test for related samples were used. Finally, a linear mixed regression model was used to evaluate the effect of optimism on functional outcomes and cytokine levels along the time, adjusted for possible confounders. Values of p less than 0.05 were considered statistically significant. To adjusting the missing data, in the linear mixed model, a subject with one-time point missing did not be dropped from the entire analysis. The remaining data were retained. No imputation procedure had been applied.

**Results**

The study group consisted of 49 subjects at baseline and 13 at 3-month follow-up. Demographic and baseline characteristics of the subjects are shown in Table 1. Most subjects were African American men who had hypertension.

Table 2 shows the sample means of each variable. The LOT-R scale was scored using total score (LOT-R), optimism subscale (OPT), and pessimism subscale (PESS) approach. The LOT-R scores in this sample were ranged from 8 to 24 with a mean of 17.3 ± 4.2. There was no significant difference in the sample mean for the LOT-R scores between men (16.6 ± 4.0) and women (18.3 ± 4.4). However, the mean of LOT-R scores among Asian (12.3 ± 2.4) was significantly lower than those among whites (17.1 ± 3.9, p=0.02) and African American (20.1 ± 3.4, p<0.001). Looking at OPT and PESS as distinct constructs, the sample means for the OPT was 9.9 ± 2.0 and 4.6 ± 3.4 for the
PESS. Similarly, no gender difference was observed in the sample means for the OPT and PESS subscales. Asian (8.0 ± 1.3, p=0.001) and Hispanics (6.9 ± 3.4, p=0.006) showed significantly higher means of the PESS subscale than African American (2.5 ± 2.8).

For specific aim 1, it was hypothesized that stroke subjects with higher levels of optimism at baseline will have lower levels of stroke severity, physical disability, and pro-inflammatory cytokines compared to those with lower optimism levels. In this study, 49 subjects were collected and reached the planned sample size for this aim. Full results of correlation analyses depicted in Table 3. As expected, the level of optimism (LOT-R) was positively correlated with OPT subscale (ρ= 0.57, p<0.001), but negatively correlated with PESS subscale (ρ= -0.88, p<0.001). LOT-R were also found negatively correlated with stroke severity (NIHSS; ρ= -0.41, p=0.003), physical disability (mRS; ρ= -0.30, p=0.05), and inflammatory markers IL-6 (ρ= -0.44, p=0.008) and CRP (ρ= -0.40, p=0.02), but not with TNF-α. Furthermore, higher scores on OPT subscale were significantly associated with lower levels of NIHSS (ρ= -0.31, p=0.03) and CRP (ρ= -0.48, p=0.004). Higher scores on PESS subscale were also significantly associated with higher levels of NIHSS (ρ= 0.33, p=0.02), mRS (ρ= 0.32, p=0.03), and IL-6 (ρ= 0.50, p=0.002). However, neither LOT-R nor OPT and PESS subscales showed a clear relation pattern with the level of TNF-α.

After performing univariate regression analyses for different outcomes on each independent variables, the variables (age, NSSQ, NIHSS, and mRS) with a p-value less than 0.1 were included in multiple linear regression models (Table 4). After adjusting the potential confounders, NIHSS and CRP were still negatively correlated with the level of
optimism. For every one unit increase of LOT-R, subject’s NIHSS scores will decrease 0.27 point (p=0.001), mRS will decrease 0.11 point (p=0.03), and level of CRP will decrease 148.6 ng/ml (p=0.02). Nevertheless, levels of IL-6 and TNF-α did not show a clear relationship with LOT-R at baseline in the regression model.

For specific aim 2, it was hypothesized that subjects’ levels of optimism would remain stable over the three months, and more optimistic subjects will have less inflammation and accelerated stroke recovery over the 3-month post-stroke period compared with less optimistic subjects. For this aim, based on two groups comparison within longitudinal data analysis, the planned sample sizes for NIHSS, mRS, and CRP levels were 24, 14, and 36, respectively. However, only 13 subjects were successfully obtained. Therefore, instead of classifying the subjects into optimistic and non-optimistic groups, the optimism scores (LOT-R) were analyzed as a continuous variable to increase the power. Based on the result in Table 5, the effect sizes of NIHSS, mRS, and CRP were 3.83, 1.75, and 2.07, respectively. Using these effect sizes, the required sample sizes for the three variables are 9, 12, 11 to achieve a power of 80% with a type I error of 0.05.

Table 5 shows the means of each variable at baseline and 3-month. Wilcoxon signed-rank test was conducted to evaluate the difference between baseline and 3-month among the 13 subjects. Levels of NIHSS, mRS, IL-6, and CRP at 3-month were significantly lower than those at baseline. TNF-α did not exhibit any significant change. Similar to the previous study (Scheier et al., 1994), levels of LOT-R, OPT, and PESS remained stable during the three-month post-stroke period.

Additionally, to adjust the informative attrition, a linear mixed model was performed to provide unbiased results. The variables LOT-R, NIHSS, mRS, IL-6, TNF-α,
and CRP, were analyzed as time-varying variables with two observations. Using
univariate linear mixed models, the results indicated that lower levels of NIHSS, mRS,
IL-6, and CRP associated with higher LOT-R scores over time. After adjusting for age,
NIHSS, and mRS, LOT-R was negatively correlated with NIHSS over three-month post-
stroke period (Table 6A, coefficient= -0.23, 95% CI: -0.36 to -0.11), but not mRS
(coefficient= -0.07, 95% CI: -0.16 to 0.01). The subjects with a higher level of LOT-R
showed lower levels of IL-6 and CRP over three-month follow-up (Table 6B,
coefficient= -0.79 and -116.5; 95% CI: -1.33 to -0.25 and -230.95 to -2.05, respectively).

Discussion

In this study, 49 ischemic stroke subjects at baseline with 13 at 3-month follow-up
visit present three key findings. First, optimism was significantly associated with reduced
stroke severity, less physical disability, and lower level of plasma CRP at baseline while
controlling for relevant medical and demographic factors. Second, subjects’ levels of
optimism remained stable over the three months. This finding parallels previous research
in college students (Scheier et al., 1994). Third, after adjusting relevant factors, optimism
was significantly associated with lower levels of IL-6 and CRP and improved stroke
severity over the 3-month post-stroke period. However, there were no significant
associations found between circulating expression level of TNF-α and optimism at
baseline and 3-month post-stroke.

The findings support recent studies on optimism and overall health in stroke
survivors. Stroke survivors with higher scores on optimism reported less physical illness
and depressive symptoms which might improve rehabilitation and recovery after stroke
(Chung, Bakas, Plue, & Williams, 2016; Mavaddat et al., 2018; Shifren & Anzaldi,
2018b). Consistently, higher levels of IL-6 and CRP were associated with lower levels of optimism in patients with cardiovascular disease and post-stroke depression (Roy et al., 2010; Wen, Weymann, Wood, & Wang, 2018).

The present study found that levels of CRP at both baseline and 3-month follow-up were significantly positively associated with the level of optimism among stroke subjects. Similarly, prior studies indicated that the level of CRP in patients with post-stroke depression (PSD) was significantly increased compared to those in patients without PSD (Cheng, Tu, Shen, Zhang, & Ji, 2018; Y. Mu, Wang, Zhou, Tan, & Wang, 2018). Additionally, ischemic stroke patients with a high CRP measured at admission associated with more severe stroke, poor functional outcome, and higher mortality (Idicula, Brogger, Naess, Waje-Andreassen, & Thomassen, 2009). At present, most of the studies reported on CRP values related prognosis and diagnosis of ischemic stroke were from the Asian population (Yu et al., 2017). However, no significant difference in the CRP levels among the ethnic groups was found in this study.

**Limitations**

There are some potential limitations to the current study. First, the sample size at the 3-month follow-up visit was relatively small (n=13). The small sample size might have decreased statistical power. However, the changes of CRP from baseline to 3-month has an effect size higher than expected and still achieve a statistical significance. Unreachable telephone number from medical records, unwilling to continue participating in the research project, and moving into nursing home were the primary factors contributed to the high attribution rate. To improve patient retention, we may (1) request for an extra contact information when consenting; (2) offer financial incentives, such as
gift cards or transport vouchers; and (3) increase patient engagement, for example, learn patients’ and caregiver’s story and share current health data through culture-centered approach (Bernstein & Feldman, 2015; Domecq et al., 2014).

Second, this was a single center study performed in an educational hospital, so the findings might not be reflective of the general stroke population. Therefore, the findings may need to be validated in a large multicenter cohort study in the future. Third, subjects with a preexisting physical disability were not excluded from this study. It might lead to misjudgment of the stroke recovery.

**Implications for Practice and Research**

According to the results, we suggest assessing patients’ level of optimism during hospitalization. Clinicians may provide an early follow-up to monitor post-stroke recovery for patients with lower level of optimism. Furthermore, an optimistic attitude can be induced by manipulating the brain’s physiological activity in a way that selectively enhances activity in one hemisphere, such as CBT, BPS, or exercise which can significantly enhance subjective and psychological well-being and reduce depressive symptoms (Bolier et al., 2013; Meevissen, Peters, & Alberts, 2011). Therefore, clinicians can recommend CBT, BPS or exercise to their patients with low levels of optimism.

Depression is characterized by overly pessimistic thought and a negative-thinking style (Hecht, 2013). Over one-third of stroke patients developed post-stroke depression (Hackett & Pickles, 2014). Therefore, future studies could longitudinally examine the relationships between optimism, depression, and coping strategies in stroke patients. Developing and evaluating appropriate psychological interventions to boost optimism may ameliorate inflammation and accelerate stroke recovery in the future. Also it would
be important to examine at which time point after stroke it would be most beneficial for patients to receive these interventions.

**Conclusions**

Stroke survivors with a higher level of optimism showed lower inflammation and better stroke recovery. A large cohort with a comprehensive analysis is necessary to determine the exact role and biological mechanism of optimism in stroke recovery. It is a worthwhile endeavor to investigate an effective therapeutic strategy by boosting the level of optimism among stroke survivors in the future.

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**Conflicts of Interest and Source of Founding**

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References


from the CoRisk study. *Neurology, 80*(14), 1278-1286.

doi:10.1212/WNL.0b013e3182887944


doi:10.1212/WNL.0000000000004922


doi:10.1586/14737175.2015.1035712


doi:10.1161/CIRCRESAHA.110.223545


doi:10.1016/j.metabol.2014.04.012

doi:10.1016/j.pneurobio.2013.11.006


doi:10.1097/MRR.0000000000000108
Moher, D. (2009). The PRISMA statement for reporting systematic reviews and
meta-analyses of studies that evaluate healthcare interventions: Explanation and
elaboration. *British Medical Journal, 339*, b2700. doi:10.1136/bmj.b2700

Luitse, M. J., van Seeters, T., Horsch, A. D., Kool, H. A., Velthuis, B. K., Kappelle, L. J.,
& Biessels, G. J. (2013). Admission hyperglycaemia and cerebral perfusion
doi:10.1159/000346588

Toyoda, K. (2012). Effect of serum lipid levels on stroke outcome after rt-PA
doi:10.1159/000334664

doi:10.1080/17439760.2016.1221122

qualitative study in the United Kingdom. *BMC Geriatrics, 18*(1), 81.

McShane, L. M., Altman, D. G., Sauerbrei, W., Taube, S. E., Gion, M., Clark, G. M., &
recommendations for tumor marker prognostic studies (REMARK). *Journal of
the National Cancer Institute, 97*(16), 1180-1184. doi:10.1093/jnci/dji237


healthcare professionals from the American Heart Association/American Stroke Association. *Stroke, 49*(3), e46-e110. doi:10.1161/STR.0000000000000158


Multi-Ethnic Study of Atherosclerosis (MESA). *Psychosomatic Medicine, 72*(2), 134-140. doi:10.1097/PSY.0b013e3181cb981b

Saver, J. L., & Altman, H. (2012). Relationship between neurologic deficit severity and final functional outcome shifts and strengthens during first hours after onset. *Stroke, 43*(6), 1537-1541. doi:10.1161/STROKEAHA.111.636928


and coagulation system activation in clinical stroke differentiation. *Journal of Neurology*, 256(1), 72-77. doi:10.1007/s00415-009-0054-8


Table 1

Demographic and characteristics of patients with ischemic stroke at baseline (n=49)

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Subjects</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years, Mean (S.D.)</td>
<td>59.29 (14.09)</td>
</tr>
<tr>
<td>Sex, male, n (%)</td>
<td>28 (57.1)</td>
</tr>
<tr>
<td>Race/ethnicity, n (%)</td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>17 (34.7)</td>
</tr>
<tr>
<td>African American</td>
<td>19 (38.8)</td>
</tr>
<tr>
<td>Hispanics</td>
<td>7 (14.3)</td>
</tr>
<tr>
<td>Asian</td>
<td>6 (12.2)</td>
</tr>
<tr>
<td>Risk factors, n (%)</td>
<td></td>
</tr>
<tr>
<td>History of stroke</td>
<td>18 (36.7)</td>
</tr>
<tr>
<td>TIA</td>
<td>1 (2.0)</td>
</tr>
<tr>
<td>Family with stroke</td>
<td>19 (38.8)</td>
</tr>
<tr>
<td>A-fib</td>
<td>7 (14.3)</td>
</tr>
<tr>
<td>Cancer</td>
<td>10 (20.4)</td>
</tr>
<tr>
<td>Cardiovascular disease</td>
<td>13 (26.5)</td>
</tr>
<tr>
<td>DM</td>
<td>24 (49.0)</td>
</tr>
<tr>
<td>HTN</td>
<td>35 (71.4)</td>
</tr>
<tr>
<td>HLD</td>
<td>26 (53.1)</td>
</tr>
<tr>
<td>Obesity</td>
<td>26 (53.1)</td>
</tr>
<tr>
<td>Depression</td>
<td>4 (8.2)</td>
</tr>
</tbody>
</table>

*Note.* S.D., Standard deviation; TIA, Transient ischemic attack; A-fib, Atrial fibrillation; DM, Diabetes mellitus; HTN, Hypertension; HLD, Hyperlipidemia.
Table 2

The levels of variables at baseline

<table>
<thead>
<tr>
<th>Variables</th>
<th>n</th>
<th>Mean ± S.D. Median (IQR)</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Optimism level</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LOT-R</td>
<td>49</td>
<td>17.31 ± 4.21</td>
<td>8 - 24</td>
</tr>
<tr>
<td>OPT</td>
<td>49</td>
<td>9.94 ± 2.05</td>
<td>6 - 12</td>
</tr>
<tr>
<td>PESS</td>
<td>49</td>
<td>4.63 ± 3.38</td>
<td>0 - 12</td>
</tr>
<tr>
<td>NIHSS</td>
<td>49</td>
<td>2.41 ± 2.42</td>
<td>0 - 9</td>
</tr>
<tr>
<td>mRS</td>
<td>47</td>
<td>2.00 (1.00 – 3.00)</td>
<td>0 - 5</td>
</tr>
<tr>
<td>IL-6, pg/ml</td>
<td>35</td>
<td>8.96 ± 8.47</td>
<td>0.53 - 27.35</td>
</tr>
<tr>
<td>TNF-α, pg/ml</td>
<td>35</td>
<td>1.96 ± 2.06</td>
<td>0.02 - 7.52</td>
</tr>
<tr>
<td>CRP, ng/ml</td>
<td>35</td>
<td>2162.34 ± 1665.16</td>
<td>78.43 - 5674.61</td>
</tr>
</tbody>
</table>

Note. S.D., Standard deviation; IQR, Interquartile range; OPT, Optimism subscale; PESS, Pessimism subscale, LOT-R, revised Life Orientation Test; NIHSS, National Institutes of Health Stroke Scale; mRS, modified Rankin Scale; IL-6, Interleukin 6; TNF-α, Tumor necrosis factor alpha; CRP, C-Reactive Protein.
Table 3

Correlation among variables in patients with ischemic stroke at baseline (n=49)

<table>
<thead>
<tr>
<th></th>
<th>OPT</th>
<th>PESS</th>
<th>LOT-R</th>
<th>NIHSS</th>
<th>mRS</th>
<th>IL-6</th>
<th>TNF-α</th>
<th>CRP</th>
</tr>
</thead>
<tbody>
<tr>
<td>OPT</td>
<td>1.000</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PESS</td>
<td>-0.135</td>
<td>1.000</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LOT-R</td>
<td>0.571**</td>
<td>-0.878**</td>
<td>1.000</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NIHSS</td>
<td>-0.312*</td>
<td>0.334*</td>
<td>-0.411**</td>
<td>1.000</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>mRS</td>
<td>-0.035</td>
<td>0.319*</td>
<td>-0.294*</td>
<td>0.158</td>
<td>1.000</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IL-6</td>
<td>-0.010</td>
<td>0.500**</td>
<td>-0.439**</td>
<td>0.227</td>
<td>0.424*</td>
<td>1.000</td>
<td></td>
<td></td>
</tr>
<tr>
<td>TNF-α</td>
<td>0.132</td>
<td>-0.218</td>
<td>0.205</td>
<td>-0.128</td>
<td>0.304</td>
<td>0.195</td>
<td>1.000</td>
<td></td>
</tr>
<tr>
<td>CRP</td>
<td>-0.476**</td>
<td>0.160</td>
<td>-0.391*</td>
<td>0.099</td>
<td>0.097</td>
<td>0.152</td>
<td>0.251</td>
<td>1.000</td>
</tr>
</tbody>
</table>

*Note. OPT, Optimism subscale; PESS, Pessimism subscale, LOT-R, revised Life Orientation Test; NIHSS, National Institutes of Health Stroke Scale; mRS, modified Rankin Scale; IL-6, Interleukin 6; TNF-α, Tumor necrosis factor alpha; CRP, C-Reactive Protein. Spearman's correlation. * p<0.05; ** p<0.01
### Table 4

*Multiple linear regression analysis for patients’ level of optimism and other clinical outcomes at baseline*

<table>
<thead>
<tr>
<th>Dependent variable</th>
<th>Independent variable</th>
<th>Estimate</th>
<th>Std. Error</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>NIHSS (n=49)</td>
<td>LOT-R</td>
<td>-0.270</td>
<td>0.070</td>
<td><strong>0.001</strong></td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>-0.030</td>
<td>0.020</td>
<td>0.240</td>
</tr>
<tr>
<td>mRS (n=47)</td>
<td>LOT-R</td>
<td>-0.110</td>
<td>0.050</td>
<td><strong>0.030</strong>*</td>
</tr>
<tr>
<td>IL-6 (n=35)</td>
<td>LOT-R</td>
<td>-0.750</td>
<td>0.390</td>
<td>0.067</td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>0.310</td>
<td>0.100</td>
<td>0.004</td>
</tr>
<tr>
<td></td>
<td>NIHSS</td>
<td>0.480</td>
<td>0.650</td>
<td>0.471</td>
</tr>
<tr>
<td></td>
<td>mRS</td>
<td>0.570</td>
<td>0.900</td>
<td>0.533</td>
</tr>
<tr>
<td></td>
<td>NSSQ</td>
<td>-0.450</td>
<td>0.410</td>
<td>0.286</td>
</tr>
<tr>
<td>TNF-α (n=35)</td>
<td>LOT-R</td>
<td>0.030</td>
<td>0.080</td>
<td>0.750</td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>0.050</td>
<td>0.030</td>
<td>0.090</td>
</tr>
<tr>
<td>CRP (n=35)</td>
<td>LOT-R</td>
<td>-148.570</td>
<td>61.770</td>
<td><strong>0.020</strong>*</td>
</tr>
</tbody>
</table>

*Note. LOT-R, revised Life Orientation Test; NIHSS, National Institutes of Health Stroke Scale; mRS, modified Rankin Scale; IL-6, Interleukin 6; TNF-α, Tumor necrosis factor alpha; CRP, C-Reactive Protein; NSSQ, Need of Support and Service Questionnaire; Std., Standard error. * p<0.05; ** p<0.01*
Table 5

The levels of variables at baseline and three-month follow-up (n=13)

<table>
<thead>
<tr>
<th>Variables</th>
<th>Baseline</th>
<th>3-month</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean ± S.D.</td>
<td>Median (IQR)</td>
<td>Mean ± S.D.</td>
</tr>
<tr>
<td></td>
<td>Range</td>
<td></td>
<td>Range</td>
</tr>
<tr>
<td>Optimism level</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LOT-R</td>
<td>15.62 ± 4.41</td>
<td>8 - 22</td>
<td>14.69 ± 5.30</td>
</tr>
<tr>
<td>OPT</td>
<td>6.62 ± 2.44</td>
<td>6 - 12</td>
<td>9.38 ± 2.53</td>
</tr>
<tr>
<td>PESS</td>
<td>6.00 ± 3.72</td>
<td>1 - 12</td>
<td>6.69 ± 3.71</td>
</tr>
<tr>
<td>NIHSS</td>
<td>3.08 ± 2.93</td>
<td>0 - 9</td>
<td>0.92 ± 2.06</td>
</tr>
<tr>
<td>mRS</td>
<td>3.00 (1.5 - 4.5)</td>
<td>0 - 5</td>
<td>2.00 (0.5 - 3.5)</td>
</tr>
<tr>
<td>IL-6, pg/ml</td>
<td>11.81 ± 8.75</td>
<td>0.76 - 25.48</td>
<td>2.76 ± 1.65</td>
</tr>
<tr>
<td>TNF-α, pg/ml</td>
<td>2.09 ± 1.77</td>
<td>0.10 - 6.29</td>
<td>1.68 ± 2.13</td>
</tr>
<tr>
<td>CRP, ng/ml</td>
<td>2530.28 ± 1857.52</td>
<td>78.43 - 5674.61</td>
<td>19.41 ± 20.67</td>
</tr>
</tbody>
</table>

Note. S.D., Standard deviation; IQR, Interquartile range; OPT, Optimism subscale; PESS, Pessimism subscale, LOT-R, revised Life Orientation Test; NIHSS, National Institutes of Health Stroke Scale; mRS, modified Rankin Scale; IL-6, Interleukin 6; TNF-α, Tumor necrosis factor alpha; CRP, C-Reactive Protein; Wilcoxon Signed-Rank Test. ** p<0.01
Table 6

*Linear mixed model analysis for patient’s level of optimism and other clinical outcomes over a three-month post-stroke period (n=13)*

**A. Stroke severity and physical disability**

<table>
<thead>
<tr>
<th>Dependent variable</th>
<th>Independent variable</th>
<th>Estimate</th>
<th>Std. Error</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>NIHSS</td>
<td>Time T2 vs T1</td>
<td>-2.25**</td>
<td>0.48</td>
<td>-3.27 to -1.24</td>
</tr>
<tr>
<td></td>
<td>LOT-R</td>
<td>-0.23**</td>
<td>0.06</td>
<td>-0.36 to -0.11</td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>-0.02</td>
<td>0.02</td>
<td>-0.06 to 0.02</td>
</tr>
<tr>
<td>mRS</td>
<td>Time T2 vs T1</td>
<td>-0.79*</td>
<td>0.28</td>
<td>-1.38 to -1.96</td>
</tr>
<tr>
<td></td>
<td>LOT-R</td>
<td>-0.07</td>
<td>0.04</td>
<td>-0.16 to 0.01</td>
</tr>
<tr>
<td></td>
<td>NIHSS</td>
<td>0.06</td>
<td>0.08</td>
<td>-0.1 to 0.22</td>
</tr>
</tbody>
</table>

**B. Pro-inflammatory cytokines**

<table>
<thead>
<tr>
<th>Dependent variable</th>
<th>Independent variable</th>
<th>Estimate</th>
<th>Std. Error</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>IL-6</td>
<td>Time T2 vs T1</td>
<td>-8.08**</td>
<td>2.37</td>
<td>-12.91 to -3.25</td>
</tr>
<tr>
<td></td>
<td>LOT-R</td>
<td>-0.79**</td>
<td>0.27</td>
<td>-1.33 to -0.25</td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>0.32**</td>
<td>0.08</td>
<td>0.16 to 0.47</td>
</tr>
<tr>
<td></td>
<td>NIHSS</td>
<td>0.55</td>
<td>0.48</td>
<td>-0.42 to 1.52</td>
</tr>
<tr>
<td></td>
<td>mRS</td>
<td>0.12</td>
<td>0.67</td>
<td>-1.26 to 1.49</td>
</tr>
<tr>
<td>TNF-α</td>
<td>Time T2 vs T1</td>
<td>-0.24</td>
<td>0.71</td>
<td>-1.69 to 1.22</td>
</tr>
<tr>
<td></td>
<td>LOT-R</td>
<td>0.01</td>
<td>0.07</td>
<td>-0.12 to 0.15</td>
</tr>
<tr>
<td>CRP</td>
<td>Time T2 vs T1</td>
<td>-2558.82**</td>
<td>553.62</td>
<td>-3684.90 to -1432.74</td>
</tr>
<tr>
<td></td>
<td>LOT-R</td>
<td>-116.5*</td>
<td>56.29</td>
<td>-230.95 to -2.05</td>
</tr>
<tr>
<td></td>
<td>NIHSS</td>
<td>-59.08</td>
<td>112.023</td>
<td>-287.07 to 168.91</td>
</tr>
</tbody>
</table>

*Note.* LOT-R, revised Life Orientation Test; NIHSS, National Institutes of Health Stroke Scale; mRS, modified Rankin Scale; IL-6, Interleukin 6; TNF-α, Tumor necrosis factor alpha; CRP, C-Reactive Protein; T1, baseline; T2, 3-month follow-up; Linear mixed model. *p<0.05; ** p<0.01
Appendix A

IRB Approval Letter

BioRepository of Neurological Diseases
NOTICE OF APPROVAL TO BEGIN RESEARCH

HSC-MH-17-0452 - BioRepository of Neurological Diseases

Number of Subjects Approved: Target: 1200 / Screen: 2000

PROVISIONS: This approval relates to the research to be conducted under the above referenced title and/or to any associated materials considered at this meeting, e.g. study documents, informed consent, etc.

APPROVED: At a Convened Meeting on 10/13/2017

EXPIRATION DATE: 09/30/2018

CHAIRPERSON: Charles C. Miller, III, PhD

Subject to any provisions noted above, you may now begin this research.

CHANGES: The principal investigator (PI) must receive approval from the CPHS before initiating any changes, including those required by the sponsor, which would affect human subjects, e.g. changes in methods or procedures, numbers or kinds of human subjects, or revisions to the informed consent document or procedures. The addition of co-investigators must also receive approval from the CPHS. ALL PROTOCOL REVISIONS MUST BE SUBMITTED TO THE SPONSOR OF THE RESEARCH.

INFORMED CONSENT DETERMINATION:
Signed Informed Consent Required

INFORMED CONSENT: Informed consent must be obtained by the PI or designee(s), using the format and procedures approved by the CPHS. The PI is responsible to instruct the designee in the methods approved by the CPHS for the consent process. The individual obtaining informed consent must also sign the consent document. Please note that only copies of the stamped approved informed consent form can be used when obtaining consent.

HEALTH INSURANCE PORTABILITY AND ACCOUNTABILITY ACT (HIPAA):

HIPAA Authorization required:
HIPAA Authorization within consent form

Waiver for Screening and Recruitment granted:
Information to be accessed: Name, Date of Birth, Medical Record Number
Information to be retained: Name, Date of Birth, Medical Record Number
UNANTICIPATED RISK OR HARM, OR ADVERSE DRUG REACTIONS: The PI will immediately inform the CPHS of any unanticipated problems involving risks to subjects or others, of any serious harm to subjects, and of any adverse drug reactions.

RECORDS: The PI will maintain adequate records, including signed consent documents if required, in a manner that ensures subject confidentiality.
Appendix B

IRB Approval Letter

Relationships among Optimism, Inflammation, and Stroke Recovery
Dr. Yun-Ju Lai  
UT-H - MS - Neurology  

June 29, 2018  

HSC-MS-18-0534 - Relationship between optimism, inflammation, and stroke recovery  

The above named project is determined to qualify for exempt status according to 45 CFR 46.101(b)  

CATEGORY #4: Research, involving the collection or study of existing data, documents, records, pathological specimens, or diagnostic specimens, if these sources are publicly available or if the information is recorded by the investigator in such a manner that subjects cannot be identified directly or through identifiers linked to the subjects.  

CHANGES: Should you choose to make any changes to the protocol that would involve the inclusion of human subjects or identified data from humans, please submit the change via iRIS to the Committee for the Protection of Human Subjects for review.  

INFORMED CONSENT DETERMINATION:  
Waiver of Consent Granted  

HEALTH INSURANCE PORTABILITY and ACCOUNTABILITY ACT (HIPAA):  
Exempt from HIPAA  

STUDY CLOSURES: Upon completion of your project, submission of a study closure report is required. The study closure report should be submitted once all data has been collected and analyzed.  

Should you have any questions, please contact the Office of Research Support Committees at 713-500-7943.
Appendix C

Revised Life Orientation Test
Revised Life Orientation Test (LOT-R)

Please respond to each question or statement by marking one box per row.

<table>
<thead>
<tr>
<th></th>
<th>Strongly disagree</th>
<th>Disagree</th>
<th>Neither agree nor disagree</th>
<th>Agree</th>
<th>Strongly agree</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>In uncertain times, I usually expect the best.</td>
<td>□</td>
<td>□</td>
<td>□</td>
<td>□</td>
</tr>
<tr>
<td>2</td>
<td>It's easy for me to relax.</td>
<td>□</td>
<td>□</td>
<td>□</td>
<td>□</td>
</tr>
<tr>
<td>3</td>
<td>If something can go wrong for me, it will.</td>
<td>□</td>
<td>□</td>
<td>□</td>
<td>□</td>
</tr>
<tr>
<td>4</td>
<td>I'm always optimistic about my future.</td>
<td>□</td>
<td>□</td>
<td>□</td>
<td>□</td>
</tr>
<tr>
<td>5</td>
<td>I enjoy my friends a lot.</td>
<td>□</td>
<td>□</td>
<td>□</td>
<td>□</td>
</tr>
<tr>
<td>6</td>
<td>It's important for me to keep busy.</td>
<td>□</td>
<td>□</td>
<td>□</td>
<td>□</td>
</tr>
<tr>
<td>7</td>
<td>I hardly ever expect things to go my way.</td>
<td>□</td>
<td>□</td>
<td>□</td>
<td>□</td>
</tr>
<tr>
<td>8</td>
<td>I don't get upset too easily.</td>
<td>□</td>
<td>□</td>
<td>□</td>
<td>□</td>
</tr>
<tr>
<td>9</td>
<td>I rarely count on good things happening to me.</td>
<td>□</td>
<td>□</td>
<td>□</td>
<td>□</td>
</tr>
<tr>
<td>10</td>
<td>Overall, I expect more good things to happen to me than bad.</td>
<td>□</td>
<td>□</td>
<td>□</td>
<td>□</td>
</tr>
</tbody>
</table>

Appendix D

National Institutes of Health Stroke Scale
**NIH Stroke Scale**

**Patient Identification:**
- Pt. Date of Birth: ____________
- Hospital: ______________________
- Date of Exam: ________________

**Interval:**
- [ ] Baseline
- [ ] 2 hours post treatment
- [ ] 24 hours post onset of symptoms ± 20 minutes
- [ ] 7-10 days
- [ ] 3 months
- [ ] Other: ____________________________(____

**Time:** __________:________ [ ]am [ ]pm

**Person Administering Scale:**

Administer stroke scale items in the order listed. Record performance in each category after each subscale exam. Do not go back and change scores. Follow directions provided for each exam technique. Scores should reflect what the patient does, not what the clinician thinks the patient can do. The clinician should record answers while administering the exam and work quickly. Except where indicated, the patient should not be coached (i.e., repeated requests to patient to make a special effort).

<table>
<thead>
<tr>
<th>Instructions</th>
<th>Scale Definition</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>1a. Level of Consciousness:</td>
<td>The investigator must choose a response if a full evaluation is prevented by such obstacles as an endotracheal tube, language barrier, orotracheal trauma and bandages. A 3 is scored only if the patient makes no movement (other than reflexive posturing) in response to noxious stimulation.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0 = Alert; keenly responsive.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1 = Not alert; but answerable by minor stimulation to obey, answer, or respond.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2 = Not alert; requires repeated stimulation to attend, or is obtunded and requires strong or painful stimulation to make movements (not stereotyped).</td>
<td></td>
</tr>
<tr>
<td></td>
<td>3 = Responds only with reflex motor or autonomic effects or totally unresponsive, flaccid, and areflexic.</td>
<td></td>
</tr>
<tr>
<td>1b. LOC Questions:</td>
<td>The patient is asked the month and his/her age.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>The answer must be correct - there is no partial credit for being close.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Aphasics and stuporous patients who do not comprehend the questions will score 2.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Patients unable to speak because of endotracheal intubation, orotracheal trauma,</td>
<td></td>
</tr>
<tr>
<td></td>
<td>severe dysarthria from any cause, language barrier, or any other problem not</td>
<td></td>
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<tr>
<td></td>
<td>secondary to aphasia are given a 1. It is important that only the initial</td>
<td></td>
</tr>
<tr>
<td></td>
<td>answer be graded and that the examiner not “help” the patient with verbal or</td>
<td></td>
</tr>
<tr>
<td></td>
<td>non-verbal cues.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0 = Answers both questions correctly.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1 = Answers one question correctly.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2 = Answers neither question correctly.</td>
<td></td>
</tr>
<tr>
<td>1c. LOC Commands:</td>
<td>The patient is asked to open and close the eyes and then to grasp and release</td>
<td></td>
</tr>
<tr>
<td></td>
<td>the non-paralytic hand. Substitute another one-step command if the hands cannot</td>
<td></td>
</tr>
<tr>
<td></td>
<td>be used. Credit is given if an unequivocal attempt is made but not completed due</td>
<td></td>
</tr>
<tr>
<td></td>
<td>to weakness. If the patient does not respond to command, the task should be</td>
<td></td>
</tr>
<tr>
<td></td>
<td>demonstrated to him or her (pantomime), and the result scored (i.e., follows</td>
<td></td>
</tr>
<tr>
<td></td>
<td>none, one or two commands). Patients with trauma, amputation, or other physical</td>
<td></td>
</tr>
<tr>
<td></td>
<td>impediments should be given suitable one-step commands. Only the first attempt</td>
<td></td>
</tr>
<tr>
<td></td>
<td>is scored.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0 = Performs both tasks correctly.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1 = Performs one task correctly.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2 = Performs neither task correctly.</td>
<td></td>
</tr>
<tr>
<td>2. Best Gaze:</td>
<td>Only horizontal eye movements will be tested. Voluntary or reflexive (oculoesophageal) eye movements will be scored, but caloric testing is not done. If the patient has a conjugate deviation of the eyes that can be overcome by voluntary or reflexive activity, the score will be 1. If a patient has an isolated peripheral nerve paresis (CN III, IV or VII), score a 2. Gaze is testable in all aphasics. Patients with ocular trauma, bandages, pre-existing blindness, or other disorder of visual acuity or fields should be tested with reflexive movements, and a choice made by the investigator. Establishing eye contact and then moving about the patient from side to side while occasionally clarifying the presence of a partial gaze palsy.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0 = Normal.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1 = Partial gaze palsy: gaze is abnormal in one or both eyes, but forced deviation</td>
<td></td>
</tr>
<tr>
<td></td>
<td>or total gaze paresis is not present.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2 = Forced deviation, or total gaze paresis not overcome by the oculoesophageal</td>
<td></td>
</tr>
<tr>
<td></td>
<td>maneuver.</td>
<td></td>
</tr>
</tbody>
</table>

Rev 10/1/2003
NIH STROKE SCALE

Interval: [ ] Baseline [ ] 2 hours post treatment [ ] 24 hours post onset of symptoms [ ] 20 minutes [ ] 7-10 days
[ ] 3 months [ ] Other ____________________________

3. Visual: Visual fields (upper and lower quadrants) are tested by confrontation, using finger counting or visual threat, as appropriate. Patients may be encouraged, but if they look at the side of the moving fingers appropriately, this can be scored as normal. If there is unilateral blindness or enucleation, visual fields in the remaining eye are scored. Score 1 only if a clear-cut asymmetry, including quadrantanopia, is found. If patient is blind from any cause, score 3. Double simultaneous stimulation is performed at this point. If there is extinction, patient receives a 1, and the results are used to respond to item 11.

4. Facial Palsy: Ask – or use pantomime to encourage – the patient to show teeth or raise eyebrows and close eyes. Score symmetry of grimace in response to noxious stimuli in the poorly responsive or non-comprehending patient. If facial trauma/bandages, ototraheal tube, tape or other physical barriers obscure the face, these should be removed to the extent possible.

5. Motor Arm: The limb is placed in the appropriate position: extend the arms (palms down) 90 degrees (if sitting) or 45 degrees (if supine). Drift is scored if the arm falls before 10 seconds. The aphasic patient is encouraged using urgency in the voice and pantomime, but not noxious stimulation. Each limb is tested in turn, beginning with the non-paraetic arm. Only in the case of amputation or joint fusion at the shoulder, the examiner should record the score as untreatable (UN), and clearly write the explanation for this choice.

6. Motor Leg: The limb is placed in the appropriate position: hold the leg at 30 degrees (always tested supine). Drift is scored if the leg falls before 5 seconds. The aphasic patient is encouraged using urgency in the voice and pantomime, but not noxious stimulation. Each limb is tested in turn, beginning with the non-paraetic leg. Only in the case of amputation or joint fusion at the hip, the examiner should record the score as untreatable (UN), and clearly write the explanation for this choice.

Patient Identification: ____________________________
Pt. Date of Birth ________________
Hospital ____________________________
Date of Exam ________________

Rev 10/1/2003
### NIH Stroke Scale

**Patient Identification:**

**Pt. Date of Birth:**

**Hospital:**

**Date of Exam:**

**Interval:**

<table>
<thead>
<tr>
<th>Option</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>[] Baseline</td>
<td></td>
</tr>
<tr>
<td>[] 2 hours post treatment</td>
<td></td>
</tr>
<tr>
<td>[] 24 hours post onset of symptoms ±20 minutes</td>
<td></td>
</tr>
<tr>
<td>[] 7-10 days</td>
<td></td>
</tr>
<tr>
<td>[] 3 months</td>
<td></td>
</tr>
<tr>
<td>[] Other</td>
<td></td>
</tr>
</tbody>
</table>

---

### 7. Limb Ataxia:

- **0 = Absent.**
- **1 = Present in one limb.**
- **2 = Present in two limbs.**
- **UN = Amputation or joint fusion, explain:**

---

### 8. Sensory:

- **0 = Normal; no sensory loss.**
- **1 = Mild-to-moderate sensory loss; patient feels pinprick is less sharp or is dull on the affected side; or there is a loss of superficial pain with pinprick, but patient is aware of being touched.**
- **2 = Severe to total sensory loss; patient is not aware of being touched in the face, arm, and leg.**

---

### 9. Best Language:

- **0 = No aphasia; normal.**
- **1 = Mild-to-moderate aphasia; some obvious loss of fluency or facility of comprehension, without significant limitation on ideas expressed or form of expression. Reduction of speech and/or comprehension, however, makes conversation about provided materials difficult or impossible. For example, in conversation about provided materials, examiner can identify picture or naming card content from patient's response.**
- **2 = Severe aphasia; all communication is through fragmentary expression; great need for inference, questioning, and guessing by the listener. Range of information that can be exchanged is limited; listener carries burden of communication. Examiner cannot identify materials provided from patient's response.**
- **3 = Mute, global aphasia; no usable speech or auditory comprehension.**

---

### 10. Dysarthria:

- **0 = Normal.**
- **1 = Mild-to-moderate dysarthria; patient slurs at least some words and, at worst, can be understood with some difficulty.**
- **2 = Severe dysarthria; patient's speech is so slurred as to be unintelligible in the absence of or out of proportion to any dysphasia, or is mute/aphonic.**

**UN = Intubated or other physical barrier, explain:**

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**Rev 10/1/2003**

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**Page 3 of 8**
Patient Identification: ___________ __________
Pt. Date of Birth __________
Hospital ________________________
________________________
Date of Exam __________

Interval: [ ] Baseline [ ] 2 hours post treatment [ ] 24 hours post onset of symptoms ± 20 minutes [ ] 7-10 days
[ ] 3 months [ ] Other __________________________

11. Extinction and Inattention (formerly Neglect): Sufficient information to identify neglect may be obtained during the prior testing. If the patient has a severe visual loss preventing visual double simultaneous stimulation, and the cutaneous stimuli are normal, the score is normal. If the patient has aphasia but does appear to attend to both sides, the score is normal. The presence of visual spatial neglect or anosagnosia may also be taken as evidence of abnormality. Since the abnormality is scored only if present, the item is never untestable.

| 0 = No abnormality. |
| 1 = Visual, tactile, auditory, spatial, or personal inattention or extinction to bilateral simultaneous stimulation in one of the sensory modalities. |
| 2 = Profound hemi-inattention or extinction to more than one modality; does not recognize own hand or orients to only one side of space. |
You know how.

Down to earth.

I got home from work.

Near the table in the dining room.

They heard him speak on the radio last night.
MAMA
TIP – TOP
FIFTY – FIFTY
THANKS
HUCKLEBERRY
BASEBALL PLAYER
Appendix E

Modified Rankin Scale
Modified Rankin Scale

<table>
<thead>
<tr>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>No symptoms at all</td>
</tr>
<tr>
<td>1</td>
<td>No significant disability despite symptoms; able to carry out all usual duties and activities</td>
</tr>
<tr>
<td>2</td>
<td>Slight disability; unable to carry out all previous activities, but able to look after own affairs without assistance</td>
</tr>
<tr>
<td>3</td>
<td>Moderate disability; requiring some help, but able to walk without assistance</td>
</tr>
<tr>
<td>4</td>
<td>Moderately severe disability; unable to walk without assistance and unable to attend to own bodily needs without assistance</td>
</tr>
<tr>
<td>5</td>
<td>Severe disability; bedridden, incontinent and requiring constant nursing care and attention</td>
</tr>
<tr>
<td>6</td>
<td>Dead</td>
</tr>
</tbody>
</table>

Total Scores (0-6): 

Appendix F

Demographic Datasheet
# Demographic datasheet

Date enrolled: __________  
Participant ID: __________

<table>
<thead>
<tr>
<th>Age: ______</th>
<th>Sex:</th>
<th>Race/Ethnicity:</th>
</tr>
</thead>
<tbody>
<tr>
<td>______</td>
<td>Male</td>
<td>___White</td>
</tr>
<tr>
<td>______</td>
<td>Female</td>
<td>___Asian</td>
</tr>
</tbody>
</table>

Medical history:

- ___Dx of Encephalopathy, delirium or confusion  
- ___Dx of Cardiac arrest  
- ___Does the patient have a history of the following?  
  - ICH  
  - Family History of Stroke  
  - History of TIA  
  - Stroke  
  - Atrial Fib  
  - Auto Immune Disease (specify which)  
  - CAD/MI w CABG/Stent  
  - CAD/MI w no CABG/Stent  
  - HTx of Cancer  
  - HTx of carotid stenosis w/CEA or Stent  
  - HTx of carotid stenosis wo/CEA or Stent  
  - CHF  
  - Coagulation Disorder  
  - Diabetes I  
  - Diabetes II  
  - Hyperlipidemia  
  - ESRD  
  - HTN  
  - Obesity  
  - Prosthetic Heart Valve  
  - PVD  
- ___Any History of Drug Abuse?  
  - Acid (LSD)  
  - Alcohol (ETOH)  
  - Amphetamines  
  - Benzodiazepines  
  - Cocaine  
  - Heroin  
  - Marijuana  
  - Methamphetamine  
  - Opiates  
  - PCP  
  - Smoker  
  - Others:
CURRICULUM VITAE
Yun-Ju Lai, PhD(c), MS, BSN, RN

EDUCATION
University of Texas, Houston, Texas 2019 PhD Nursing
University of Texas, Houston, Texas 2015 BSN Nursing
National Tsing Hua University, Hsinchu, Taiwan 2002 MS Life Science
Fu Jen Catholic University, Taipei, Taiwan 2000 BS Biology

PROFESSIONAL POSITIONS
The University of Texas Science Center at Houston, McGovern Medical School, Department of Neurology, Houston, TX (PI: Louise McCullough, MD/PhD) Graduate Assistant 2016 - Present
Memorial Hermann-Texas Medical Center Houston, Texas Clinical Nurse 2015 - 2018
The University of Texas M. D. Anderson Cancer Center, Department of Experimental Therapeutics, Houston, TX (PI: Bryant G. Darnay, PhD) Research Assistant 2010 – 2012
Vanung University, Department of Biotechnology, Taoyuan, Taiwan Teaching Instructor 2004 - 2008
Chung Yuan Christian University, Department of Bioscience Technology, Taoyuan, Taiwan Instructor 2002 - 2004

PROFESSIONAL MEMBERSHIPS
Sigma Theta Tau International Society of Nursing 2018 - Present
Southern Nursing Research Society 2018 - Present
The Council for the Advancement of Nursing Science 2018 - Present
American Heart Association 2017 - Present

Chinese Nursing Association of Houston 2015 - Present

Cizik UTHealth School of Nursing Student Government Organization 2014 - Present

**PUBLICATIONS**

**Research Articles:**


**Book Chapter**

**Press Release:**
1. Better living through better thinking. Out in Front Reports. 2018 November, Brain and behavioral health. [https://www.uth.edu/out-in-front/story.htm?id=6a02ce3-a71c-4420-855e-55ee69a6c0b9](https://www.uth.edu/out-in-front/story.htm?id=6a02ce3-a71c-4420-855e-55ee69a6c0b9)

**PRESENTATIONS**

**Oral Presentations**
1. “Relationships among optimism, inflammation, and stroke recovery” Stroke Transitions Education and Prevention (STEP), The University of Texas Science Center at Houston, McGovern Medical School, Department of Neurology, Houston, TX (Apr. 2019)
2. “Nursing education and careers in the U.S.” The sixth affiliated hospital of Sun Yat-Sen University GuangDong Gastrointestinal Hospital, China (Nov. 2018)

**Poster Presentations**


**HONORS AND AWARDS**

UTHealth School of Nursing, Research Day Best Poster Award, 2017

UTHealth School of Nursing, Ph.D. Nursing Student Best Poster, 2017

Crawford and Hattie Jackson Foundation Scholarship ($5,000), 2016

Fu Jen Catholic University, Taipei, Taiwan, The second place honor, Spring Semester 2000

National Tsing Hua University, Hsinchu, Taiwan, Recommendation-Selection Admission Program, 1999

Fu Jen Catholic University, Taipei, Taiwan, The second place honor, Fall Semester 1999

Fu Jen Catholic University, Taipei, Taiwan, The second place honor, Spring Semester 1999

**CURRENT GRANT SUPPORT**

Summer 2018 Predoctoral Fellowship (Award Number: 18PRE34060017)

Title: PD-L1 mediated T cell infiltration across the blood-brain barrier in aging and stroke

Agency: American Heart Association (AHA) (PI: Yun-Ju Lai)

Amount: $ 53,688 Total

07/01/2018 - 06/30/2020