### General Information

<table>
<thead>
<tr>
<th>Protocol:</th>
<th>AAAR1041(M00Y01)</th>
<th>Protocol Status:</th>
<th>Approved</th>
</tr>
</thead>
<tbody>
<tr>
<td>Effective Date:</td>
<td>02/09/2017</td>
<td>Expiration Date:</td>
<td>01/24/2018</td>
</tr>
<tr>
<td>Originating Department Code:</td>
<td>MED Molecular Medicine (751940X)</td>
<td>Principal Investigator:</td>
<td>Wang, Nan (nw30)</td>
</tr>
<tr>
<td>From what Columbia campus does this research originate:</td>
<td>Medical Center</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Title:</td>
<td>Impact of LDL-cholesterol lowering on platelet activation</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Protocol Version #:</td>
<td>1</td>
<td>Abbreviated Title:</td>
<td>Impact of LDL-cholesterol lowering on platelet activation</td>
</tr>
</tbody>
</table>

Was this protocol previously assigned a number by an IRB: No

Is the purpose of this submission to obtain a "Not Human Subjects Research" determination? No

### Attributes

Special review type: Check all that apply or check "None of the Above" box.
- [ ] Review for 45 CFR 46.118 Determination (involvement of human subjects is anticipated but is not yet defined)
- [ ] Funding review for Administrative IRB approval (such as for Center or Training Grants)
- [x] None of the above

IRB of record information: Will a Columbia IRB be the IRB that is responsible for providing review, approval, and oversight for this study?
- Yes
  - Select the most appropriate response:
    - Columbia will be the IRB of record for the study procedures conducted by Columbia researchers (Note: this response will apply to most submissions).

Is this research part of a multicenter study?
- No

Please indicate if any of the following University resources are utilized:
- [ ] Cancer Center Clinical Protocol Data Management Compliance Core (CPDM)
- [ ] CTSA-Irving Institute Clinical Research Resource (CRR)
- [ ] CTSA- Irving Institute Columbia Community Partnership for Health (CCPH)
- [x] None of the above

### Background

Abbreviated Submission:
The IRB has an abbreviated submission process for multicenter studies supported by industry or NIH.
cooperative groups (e.g., ACTG, HVTN, NCI oncology group studies, etc.), and other studies that have a complete stand-alone protocol. The process requires completion of all Rascal fields that provide information regarding local implementation of the study. However, entering study information into all of the relevant Rascal fields is not required, as the Columbia IRBs will rely on the attached stand-alone (e.g., sponsor’s) protocol for review of the overall objectives.

If you select the Abbreviated Submission checkbox and a section is not covered by the attached stand-alone protocol, you will need to go back and provide this information in your submission.

Study Purpose and Rationale:
Provide pertinent background description with references that are related to the need to conduct this study. If this is a clinical trial, the background should include both preclinical and clinical data. Be brief and to the point.

[ ] Abbreviated Submission - This information is included in an attached stand-alone protocol. Proceed to the next question

Hyperlipidemia as exemplified by familial hypercholesterolemia is associated with increased platelet activation and an underlying pro-coagulant state\(^1\)\(^-\)\(^3\). Hyperlipidemia primes platelets and increases platelet activation in response to various agonists\(^1\), \(^4\). Plasma cholesterol levels appear to have a critical role in modulating platelet activity as hypercholesterolemia increases platelet activation more potently than hypertriglyceridemia\(^1\), \(^4\). Increased platelet reactivity may contribute to the increased risk of atherothrombosis associated with hypercholesterolemia. Plasma levels of platelet activation markers such as thrombin-antithrombin complex (TAT), soluble P-selectin (sP-selectin), soluble CD40L (sCD40L) or P-selectin exposure at surface of platelets are increased in hypercholesterolemic patients\(^5\)\(^-\)\(^7\). Increased levels of the platelet activation markers are associated with increased platelet membrane cholesterol content in hypercholesterolemia\(^8\). Statins may show antithrombotic properties\(^9\). The PROVE-IT trial, a study to assess the impact of statins on acute coronary syndrome (ACS), demonstrated an early beneficial effect after 30 days of follow-up that significantly reduced the risk of death, MI or rehospitalization\(^10\). Additional human studies suggest that the benefits may come partly from the antithrombotic properties of statins\(^9\). Statins reduced the level of TAT, sP-selectin, sCD40L and P-selectin exposure at surface of platelets\(^8\), \(^11\)\(^-\)\(^13\). These studies suggest an early direct effect of statins on platelets and a late prolonged effect that is associated with progressive reduction of plasma LDL-cholesterol levels\(^9\), \(^14\). Both the early and late effects are proposed to lower platelet activation and contribute to the reduced risk.

Human genome wide association studies have revealed novel genetic loci associated with coronary heart disease. One such locus resides in LNK/SH2B3 which in mice is expressed in hematopoietic cells and suppresses thrombopoietin signaling via its receptor MPL. Using human cord blood, we recently showed that the common TT risk genotype (R262W) of LNK is associated with expansion of hematopoietic stem cells and enhanced megakaryopoiesis, demonstrating reduced LNK function and increased MPL signaling. To model the human reduced LNK function associated with the TT risk genotype, we used Lnk\(^\wedge\)\(^-\)\(^/-\) mice. In mice, hematopoietic Lnk deficiency led to increased platelet activation and accelerated arterial thrombosis, but only in the setting of hypercholesterolemia. Increased platelet activation combines with hypercholesterolemia induced myelopoiesis to promote pro-thrombotic platelet/leukocyte aggregate formation and accelerated atherogenesis in LNK deficiency. In light of these findings, we propose the following studies as an investigator initiated study. The primary goal is to assess the impact of Evolocumab therapy on platelet function of familial hypercholesterolemia (FH) patients in a randomized, double blind study. Evolocumab is a humanized monoclonal antibody that targets circulating PCSK9, increases hepatic LDL receptor, decreases plasma LDL cholesterol and reduces risk of cardiovascular events\(^15\), \(^16\). Evolocumab (brand name Rapatha) has been approved by FDA along with diet and maximally tolerated statin therapy in adults with FH or atherosclerotic heart or blood vessel problems, who need additional lowering of LDL cholesterol. The secondary goal is to determine if platelet activation or the response to Evolocumab therapy is modified by
rs3184504 polymorphism. We believe that these investigations will complement ongoing studies to demonstrate that Evolocumab reduces athero-thrombotic risk and aid the decision-making as to whether Evolocumab can reduce the atherothrombotic risk in ACS patients.


**Study Design:**
Describe the methodology that will be used in this study, covering such factors as retrospective vs. prospective data collection, interventional vs. non-interventional, randomized vs. non-randomized, observational, experimental, ethnography, etc.

[x] Abbreviated Submission - This information is included in an attached stand-alone protocol. Proceed to the next question
Statistical Procedures:
Provide sufficient details so that the adequacy of the statistical procedures can be evaluated including power calculations to justify the number of participants to be enrolled into the study. Definitions of subject terms such as enrolled and accrued as used for Rascal submissions can be found in the Subjects section.

Abbreviated Submission - This information is included in an attached stand-alone protocol. Proceed to the next question

The effect of PCSK9 inhibitors, including evolocumab, on platelet activation measures has not been assessed previously. However, our hypothesis is based on the LDL-cholesterol lowering effect of evolocumab, and evolocumab has been demonstrated to have at least as potent an effect on LDL lowering as statins\(^1\),\(^2\). Statin therapy in hypercholesterolemia patients results in 50% reduction in LDL-cholesterol levels and add-on evolocumab therapy indicated an additional 50-60% reduction\(^1\),\(^2\). Therefore, we base our power estimates on platelet activation data available for statins. **Primary measure/platelet aggregation** - The primary measurement will be thrombin receptor activating peptide-stimulated platelet aggregation as assessed by the commercially-available VerifyNow P2Y12 assay (Accriva Diagnostics). The assay read-outs are 1) PRU, platelet reactivity units (specific for P2Y12 receptors); 2) BASE, total platelet function despite P2Y12 receptor blockage; and 3) IPA, percent platelet P2Y12 inhibition calculated as [(BASE - PRU) / BASE] X 100. Two reports involving patients on antiplatelet therapy with high residual platelet reactivity demonstrated an effect of statins on lowering platelet aggregation via the VerifyNow assay. In CAD patients treated with clopidogrel, the addition of atorvastatin for 30 days resulted in an absolute mean difference of 102 ± 54 PRU compared to baseline, corresponding to a 37 ± 14% IPA\(^5\). In a separate study of CAD/diabetic patients treated with clopidogrel, the addition of atorvastatin for 30 days resulted in an absolute mean difference of 106 ± 75 PRU compared to baseline, corresponding to a 33 ± 20% IPA\(^6\). Similar effect sizes and variabilities of statins on reductions in platelet aggregation were observed in type II hyperlipidemic patients using light transmittance aggregometry\(^7\). The reference range for the VerifyNow assay in individuals not on antiplatelet therapy is 180-376 PRU. These data are continuous and normally distributed\(^5\),\(^6\). Thus, means and standard deviations will be used to describe reductions in platelet aggregation. Based on reported literature indicating additional effects of statins on platelet function following 30-day vs shorter periods of treatment, we have revised our experimental design to have a primary endpoint of 30-day treatment\(^3\),\(^7\). Within-group thirty day evolocumab treatment will be tested by one-group paired t-test (two-sided) using each subject as its own control (evolocumab treatment vs vehicle pre-treatment control). Between-group thirty day evolocumab treatment will be tested by standard two-group t-test (two-sided, evolocumab vs vehicle control group run in parallel). The sample size estimates have been run using one-group, paired t-test published data. Using PRU mean difference = 102\(^3\), SD = 54\(^3\), power = 0.8 and alpha = 0.05, we estimate 6 subjects. Since the effects of statins on platelet aggregation may be mediated by both LDL-lowering and non-LDL-lowering mechanisms, we can calculate the sample size based on 50% reduced mean difference (PRU = 51). Then, we estimate 11 subjects. An equal number of subjects will be required for the placebo group run in parallel. Target enrollment will include all patients screened; we estimate that 10% may not be eligible based on exclusion criteria and/or may not return for scheduled blood draws. Thus, for 80% power, the target enrollment is 24 subjects over two years. **Secondary measure/platelet P-selectin exposure** – Secondary measures will include various cellular and circulating markers of platelet activation measured by flow cytometry or commercially-available enzyme-linked immunoassays (ELISAs). The target enrollment of 24 subjects should provide at least 80% power to detect differences between treatment groups based on data reported in the literature. For example, platelet P-selectin level measured by flow cytometry and reported as % positive cells (out of 50,000 cells counted) showed a mean reduction ± SD of 10.9 ± 6.0 (simvastatin). \(10.9 ± 5.6\)
(atorvastatin), 7.2 ± 6.8 (fluvastatin) and 9.0 ± 6.8 (pravastatin) following 30 days of statin treatment in hypercholesterolemic patients compared with untreated controls. In a separate study of stroke patients with hypercholesterolemia, platelet P-selectin showed a mean reduction of 41 ± 27% positive cells in response to thrombin following 6 months of treatment with simvastatin compared to baseline measurements in the same subjects before statin treatment. An additional effect of simvastatin treatment was observed for soluble P-selectin serum concentrations measured by ELISA. Soluble P-selectin showed a mean reduction of 46.4 ± 49 ng/ml compared to baseline measurements in the same subjects before statin treatment. The design of this study makes it appropriate to use paired t test for data analysis.

References
Is the purpose of this submission to seek expedited review, as per the federal categories referenced in 45CFR46.110?
No

### Funding

Is there any external funding or support that is applied for or awarded, or are you the recipient of a gift, for this project?
Yes

<table>
<thead>
<tr>
<th>Award Type</th>
<th>Funding Source Name</th>
<th>Name of awarding agency</th>
<th>Status</th>
<th>Award # or Application Date</th>
<th>Federal/State/Local Government Direct or Subcontract</th>
<th>What is the award covering?</th>
<th>Rascal PT Number</th>
</tr>
</thead>
<tbody>
<tr>
<td>Industry</td>
<td>Amgen</td>
<td></td>
<td></td>
<td></td>
<td>Entire Protocol</td>
<td></td>
<td>PT-AABN1794</td>
</tr>
</tbody>
</table>

### Locations

<table>
<thead>
<tr>
<th>Location Type</th>
<th>Facility Name</th>
<th>Domestic or International</th>
<th>Geographic Location</th>
<th>Local IRB Ethics Approval</th>
<th>Local Site Approval</th>
</tr>
</thead>
<tbody>
<tr>
<td>NewYork-Presbyterian Hospital @ Columbia</td>
<td>622 West 168 Street, PH 10-305</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Personnel

<table>
<thead>
<tr>
<th>UNI</th>
<th>Name</th>
<th>Role</th>
<th>Department</th>
<th>Edit/View</th>
<th>Obtaining Informed Consent</th>
</tr>
</thead>
<tbody>
<tr>
<td>nw30</td>
<td>Wang, Nan</td>
<td>Principal Investigator</td>
<td>MED Molecular Medicine (751940X)</td>
<td>Edit</td>
<td>Y</td>
</tr>
<tr>
<td>cbw13</td>
<td>Welch, Carrie</td>
<td>Investigator</td>
<td>MED Molecular Medicine (751940X)</td>
<td>Edit</td>
<td>N</td>
</tr>
<tr>
<td>hng1</td>
<td>Ginsberg, Henry</td>
<td>Investigator</td>
<td>MED Preventive Medicine (751920X)</td>
<td>Edit</td>
<td>Y</td>
</tr>
<tr>
<td>lao2106</td>
<td>Oling, Lilian</td>
<td>Non-Engaged Personnel</td>
<td>MED Molecular Medicine (751940X)</td>
<td>Edit</td>
<td>N</td>
</tr>
</tbody>
</table>

**Roles and Experience:**
- Principal Investigator. Over 30 years of research experience.
- Investigator. Over 30 years of research experience.
- Investigator. Over 40 years of basic and clinical research experience.
- Administrator.
UNI | Name | Role | Department | Edit/View | Obtaining Informed Consent
--- | --- | --- | --- | --- | ---
lt2453 | Tascau, Liana | Other Engaged Personnel | MED Molecular Medicine (751940X) | View | N

Roles and Experience: Research staff. Over 20 years of research experience.

mc1336 | Cruz, Marisol | Non-Engaged Personnel | MED Molecular Medicine (751940X) | Edit | N

Roles and Experience: Administrator.

ww2351 | Wang, Wei | Investigator | MED Immunology (751910X) | Edit | N

Roles and Experience: Investigator. Over 5 years of research experience.

---

**Training and COI**

The PI must ensure that each individual that is added as personnel has met the training requirements for this study (http://www.cumc.columbia.edu/dept/irb/education/index.html). For help identifying which research compliance trainings you may be required to take, visit the Research Compliance Training Finder.

---

### Training and COI Table

<table>
<thead>
<tr>
<th>UNI</th>
<th>Name</th>
<th>COI</th>
<th>HIPAA</th>
<th>HSP (CITI)</th>
<th>Research with Minors (CITI)</th>
<th>FDA-Regulated Research (CITI)</th>
<th>S-I</th>
<th>CRC</th>
<th>Good Clinical Practice (GCP)</th>
<th>Genetic Research Consent</th>
</tr>
</thead>
<tbody>
<tr>
<td>nw30</td>
<td>Wang, Nan</td>
<td>09/06/2016</td>
<td>11/10/2016</td>
<td>11/18/2016</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>cbw13</td>
<td>Welch, Carrie</td>
<td>03/02/2016</td>
<td>11/10/2016</td>
<td>04/03/2016</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>hng1</td>
<td>Ginsberg, Henry</td>
<td>10/05/2016</td>
<td>05/31/2014</td>
<td>03/31/2014</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>lao2106</td>
<td>Oling, Lilian</td>
<td>05/24/2016</td>
<td>01/07/2018</td>
<td>07/18/2016</td>
<td>08/02/2011</td>
<td>08/02/2011</td>
<td></td>
<td>01/06/2011</td>
<td></td>
<td></td>
</tr>
<tr>
<td>lt2453</td>
<td>Tascau, Liana</td>
<td>04/24/2016</td>
<td>01/04/2015</td>
<td>09/02/2014</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>mc1336</td>
<td>Cruz, Marisol</td>
<td>09/08/2016</td>
<td>01/24/2017</td>
<td>09/02/2014</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

---

**Departmental Approvers**

Electronic Signature: Liana Tascau (751940X) - Other Engaged Personnel  
Date: 11/29/2016

Electronic Signature: Henry Ginsberg (751920X) - Investigator  
Date: 11/29/2016

Electronic Signature: Carrie Welch (751940X) - Investigator  
Date: 11/29/2016

Electronic Signature: Alan Tall (751940X) - Director  
Date: 11/29/2016

Electronic Signature: Nan Wang (751940X) - Principal Investigator  
Date: 02/07/2017

Electronic Signature: Wei Wang (751910X) - Investigator  
Date: 11/29/2016

---

**Privacy & Data Security**
Indicate the methods by which data/research records will be maintained or stored (select all that apply):

- [ ] Hardcopy (i.e., paper)
- [x] Electronic

Where will the data be stored?

- [ ] On a System
- [x] On an Endpoint

Identify what type of endpoint will be used (select all that apply):

- [ ] Desktop Computer
- [x] Laptop Computer
- [ ] Mobile Device
- [ ] Other

Does this study involve the receipt or collection of Sensitive Data?

Yes

If any Sensitive Data is lost or stolen as part of your research protocol, you must inform both the IRB and the appropriate IT Security Office (CUMC IT Security if at CUMC; CUIT if at any other University campus).

What type of Sensitive Data will be obtained or collected? Select all that apply:

- [ ] Personally Identifiable Information (PII), including Social Security Numbers (SSN)

Will Social Security Numbers (SSNs) be collected for any purpose?

- [x] Protected Health Information (PHI), including a Limited Data Set (LDS)

If any PHI is lost or stolen, you must inform both the IRB and the Office of HIPAA Compliance.

Indicate plans for secure storage of electronic sensitive data: check all that apply

- [ ] Sensitive data will not be stored in electronic format
- [ ] Sensitive data will be stored on a multi-user system
- [x] Sensitive data will be stored on an encrypted endpoint

By Selecting an Endpoint Device and approving this protocol for submission to the IRB, the PI is attesting that the device and any removable media that may be used have been or will be registered and/or will be maintained in compliance with the University's Information Security Charter and all related policies. It is important that this information is updated, during the course of the study, as new devices are added.

Provide a description of how the confidentiality of study data will be ensured, addressing concerns or protections that specifically relate to the data storage elements identified above (e.g. hard copy, electronic, system, and/or endpoint):

Any information from this study will remain confidential. All data and information generated from the study will be stored in encrypted, password protected university or hospital computers. All sensitive data will be stored in Crown and iNYP systems of CUMC. All personal identifiers, including medical record number, social security number, subject name an initials, phone numbers and addresses, will not be used as coding mechanism. A unique alphanumeric code number will be assigned to each study subject in order to protect his or her identity and maintain confidentiality. Subject source documents containing identifying information, such as the signed consent form as well as the key linking the unique alphanumeric number to subject identity, will be separately and securely stored in paper form in a locked filing cabinet in a...
locked office at CUMC. Coded data will be securely stored in electronic form on a fully encrypted password protected computer, located in a locked office at CUMC. Data will be accessible only to a limited number of key study team members. The coded data will not be de-identified.

**Is there or will there be a Certificate of Confidentiality (CoC) for this research?**  
No

---

**Provide a description of the protections in place to safeguard participants’ privacy while information is being collected:**

Any information from this study will remain confidential. The consent form discloses to potential participants the individuals and/or agencies that are able to look at and copy research records, namely:
- the investigator, study staff and other medical professionals who may be evaluating the study;
- authorities from CUMC, including their IRB committee.

Information made available for inspection will be handled in strict confidence and in accordance with local data protection laws.

Note that the sensitive data containing personal identifiers are not required data to test the hypothesis of this study in order to achieve the stated goals. Therefore, these sensitive data will be securely maintained and stored by Dr. Henry Ginsberg and staff of Research Pharmacy of CUMC who directly contact the participants. Other investigators of this study who do not directly contact the participants but are involved in sample assays and data analysis will have access only to the coded data.

---

**Procedures**

**Is this project a clinical trial?**  
Yes

**Is this project a clinical trial that requires registration with www.clinicaltrials.gov?**  
Yes

**Has this study been registered with www.clinicaltrials.gov?**  
No

Please note that this section should be updated when the registration number is received. At this time, please indicate who will be responsible for registering the study:

Nan Wang

**Is this project associated with, or an extension of, an existing Rascal protocol?**  
No

**Do study procedures involve any of the following?**

- Analysis of existing data and/or prospective record review  
  No
- Audio and/or video recording of research subjects  
  No
- Behavioral Intervention?  
  No
- Biological specimens (collection or use of)  
  Yes
- Cancer-related research  
  No
- Drugs or Biologics
Yes
Future use of data and/or specimens
Yes
Genetic research
Yes

Indicate which, if any, of the following apply:

Genetic Testing as defined by NYS 79-l
Yes

Will the results of the genetic test be given to the subjects and/or placed in their medical records?
No

Gene Transfer
No

Generation of large scale genomic data (e.g. GWAS studies)
No

Human embryos or human embryonic stem cells
No

Imaging procedures or radiation
No

Medical Devices
No

Surgical procedures that would not otherwise be conducted or are beyond standard of care
No

Will any of the following qualitative research methods be used?

Survey/interview/questionnaire
No

Systematic observation of public or group behavior
No

Program evaluation
No

Will any of the following tests or evaluations be used?

Cognitive testing
No

Educational testing
No

Non-invasive physical measurements
No

Taste testing
No

Is there an external protocol that describes ALL procedures in this study?
Yes

[x] Check here if all procedures being conducted by Columbia researchers are detailed in the stand-alone protocol, or provide a detailed description of which procedures are being conducted by Columbia researchers.

---

Biological Specimens

Add an individual entry for each human specimen type that will be collected or utilized for the proposed study.
For each specimen type, indicate the source or sources from which you will obtain the specimens.

The use of specimens for research purposes may require that informed consent (or a waiver, if applicable) and HIPAA Authorization (or a waiver, if applicable) be obtained from subjects.

Type: Blood

Source:

[x] From Columbia and/or NYP Subjects/Patients or Repositories managed by Columbia

Select all that apply to this specimen type:

[x] Specimens will be prospectively collected specifically for this research.

[ ] Residual specimens from clinical care that would otherwise be discarded have been or will be collected.

[ ] Specimens to be analyzed will be (or have been) collected from a commercial source.

[ ] Specimens to be analyzed will be (or have been) collected under a separate CU IRB-approved protocol; this could be an approved research repository protocol.

[ ] From Non-Columbia/NYP Subjects/Patients or Repositories not managed by Columbia

Description of Specimen and Method of Obtaining:

Blood samples. Blood (18 ml) will be collected by venipuncture in the morning from the resting and fasting subjects using a G21 bufferfly needle, as detailed in the protocol.

Indicate the manner in which the specimens will be labeled:

[ ] Specimens will be labeled with direct identifiers

[x] Specimens will be labeled with a code and the research team has the key and can link specimens to direct identifiers. This code would be considered an indirect identifier

[ ] The identifiers will be removed prior to the receipt of the specimens by Columbia researchers and no link will remain

[ ] Specimens were originally collected without identifiers

If specimens are collected or received at any point in time as with direct or indirect identifiers by the current researchers, then the specimens are considered to be identifiable, and the requirements for Informed Consent (or a waiver, if applicable) and HIPAA Authorization (or a waiver, if applicable) apply. The necessary information will need to be included in the respective sections of this Rascal submission.

Drugs/Biologics

On the General Information page you have indicated that the protocol version associated with the use of this drug/biologic is as follows: 1

Please note that a Protocol Version # is required for protocols using a drug or biologic, and you will not be allowed to submit this protocol until the Protocol Version # field is complete. Please ensure that the Protocol Version # is completely and accurately reported on the General Information page.

List each drug or biologic that will be administered as the object of the protocol or is being used because it is relevant to the aims of the research protocol. This applies whether the drug/biologic is not yet FDA-approved (i.e., is investigational), is FDA approved and used in accordance with its labeling, or is an approved product that is being used in an investigational manner (i.e., off-label use is being studied).

Note that the questions apply only to drugs used in clinical investigations. Emergency use of a drug that is not
yet FDA-approved is not a clinical investigation, and a submission in Rascal may not be required. Please contact the IRB for assistance if emergency use of a drug or biologic that is not yet FDA-approved is being considered: (212) 305-5883.

Name: Evolucumab
Dose: 140mg
Study phase: Other
   Please Explain:
   The drug has already been approved by the FDA

Manufacture Information

Name: Amgen
Address: One Amgen Center Drive, Thousand Oaks, CA 91320-1799
Contact Information: Irma Lopez
   Manager, Non-Amgen Sponsored Clinical Research (NASCER)
   Global Clinical Program Management
   A
   One Amgen Center Drive, MS 38-3-B
   Thousand Oaks, CA 91320
   805-447-3769
   626-808-3034 (Mobile)
   lopezir@amgen.com

Route of administration: Subcutaneous
Is the drug/biologic FDA-approved and used in accordance with its labeling? Yes
An IND/BB-IND is not required. A copy of the package insert must be attached.

Future Use

For what materials do you anticipate future research use? Select all that apply.
   [x] Data
   [x] Biological Specimens

For what materials do you anticipate future research use? Select all that apply.
   [x] Some or all data and/or specimens, as applicable, will be retained by Columbia researchers for future use.

   How are the materials intended to be used for research in the future?
   Current PI will retain the materials and there is no intent to create a repository or share with other CU researchers.
   Note: Information provided in original consent forms will be will be considered when an addition of future uses is submitted via modification.

   What future uses are anticipated?
   Additional or new plasma surrogate markers may need to be assessed in the future in association with Evolucumab therapy.

   How will the data and/or specimens, as applicable, be labeled during storage for future uses.
   [x] In the same manner as during collection (e.g., with direct identifiers, coded, de-identified, anonymous)
   [ ] In a different manner than during collection. Select all that apply:

Describe the physical storage for the specimens/data, including location.
In the same manner as during collection
[x] In a different manner than during collection

**Describe the physical storage.**

The blood and plasma samples will be stored in -80°C freezer securely located in PS 8-401

**Describe who will have access to the stored data and/or specimens.**

Nan Wang, Henry Ginsberg

[ ] Some or all data/specimens will be released to a non-Columbia entity for future use and Columbia researchers will not have direct control.

---

**Recruitment And Consent**

**Recruitment:**

Describe how participants will be recruited:

All adult male and female subjects who have a clinical diagnosis of hypercholesterolemia and who are referred to Dr. Ginsberg’s Lipid Practice for treatment with PCSK9 inhibitor, and with LDL cholesterol levels >100 mg/dl on baseline treatment with statins and/or ezitimibe will be recruited for the study.

Select all methods by which participants will be recruited:

- [x] Person to Person
- [ ] Radio
- [ ] Newspapers
- [ ] Direct Mail
- [ ] Website
- [ ] Email
- [ ] Television
- [ ] Telephone
- [ ] Flyer/Handout
- [ ] Newsletter/Magazine/Journal
- [ ] ResearchMatch
- [ ] CUMC RecruitMe

**Informed Consent Process:**

**Informed Consent Process, Waiver or Exemption:** Select all that apply

- [x] Informed consent with written documentation will be obtained from the research participant or appropriate representative.

  **Documentation of informed consent is applicable to:**

  The study in its entirety

  Identify the portion of the study (e.g., prospective portion, focus groups, substudy 2) or subject population for which documentation of consent will be obtained:
Documentation of participation will be obtained from:

- [x] Adult participants
- [ ] Parent providing permission for a child's involvement
- [ ] Legally Authorized Representatives (LARs)

Describe how participants' written consent will be obtained:
Consent will be obtained using a signed consent form. The Investigator will explain the nature of the study to the participants. Consent form will be administered and signed by both the participant and the investigator.

Informed consent is not required for exempt research but is recommended for such research when there will be interaction with research participants for the purpose of the research.

- [ ] Informed consent will be obtained but a waiver of written documentation of consent (i.e., agreement to participate in the research without a signature on a consent document) is requested.
- [ ] A waiver of some or all elements of informed consent (45 CFR 46.116) is requested.
- [ ] Planned Emergency Research with an exception from informed consent as per 21 CFR 50.24.
- [ ] Informed consent is not required; this is exempt research.

Subject Language
Enrollment of non-English speaking subjects is not expected.

During the course of the study, if non-English speaking subjects are encountered, refer to the IRB's policy on the Enrollment of Non-English Speaking Subjects in Research for further details (http://www.cumc.columbia.edu/dept/irb/policies/documents/Nonenglishspeakingsubjects.Revised.FINALDRAFT.111909.website.doc)

Capacity to Provide Consent:
Do you anticipate using surrogate consent or is research being done in a population where capacity to consent may be questionable?
No

Research Aims & Abstracts

Research Question(s)/Hypothesis(es):
We hypothesize that platelet activation is heightened in hypercholesterolemia by cholesterol enrichment of platelets. This is reversed by Evolucumab in association with reduced plasma LDL cholesterol levels. We also hypothesize that platelet reactivity in hypercholesterolemia and in response to Evolucumab therapy is modified by rs3184504 polymorphism. TT risk allele increases platelet reactivity in hypercholesterolemia which is reversed by Evolucumab.
Scientific Abstract:

Hyperlipidemia as exemplified by familial hypercholesterolemia is associated with increased platelet activation and an underlying pro-coagulant state. Hyperlipidemia primes platelets and increases platelet activation in response to various agonists. Plasma cholesterol levels appear to have a critical role in modulating platelet activity as hypercholesterolemia increases platelet activation more potently than hypertriglyceridemia. Plasma levels of platelet activation markers such as thrombin-antithrombin complex (TAT), soluble P-selectin (sP-selectin), soluble CD40L (sCD40L) or P-selectin exposure at surface of platelets are increased in hypercholesterolemic patients. Increased levels of the platelet activation markers are associated with increased platelet membrane cholesterol content in hypercholesterolemia. Statins may show antithrombotic properties. The PROVE-IT trial, a study to assess the impact of statins on acute coronary syndrome (ACS), demonstrated an early beneficial effect after 30 days of follow-up that significantly reduced the risk of death, MI or rehospitalization. A retrospective study of 1616 patients (PRISM) also showed significant reduction of the risk during 30 day follow-up in ACS patients with statin therapy. Additional human studies suggest that the benefits may come partly from the antithrombotic properties of statins. Statins reduced the level of TAT, sP-selectin, sCD40L and P-selectin exposure at surface of platelets. Reduced levels of P-selectin at the platelet surface were associated with reduced platelet cholesterol content in subjects treated with statins. These studies suggest an early direct effect of statins on platelets and a late prolonged effect that is associated with progressive reduction of plasma LDL-cholesterol levels. Both the early and late effects are proposed to lower platelet activation and contribute to the reduced risk. Consistent with the idea that cholesterol enrichment increases platelet activation, infusions of a reconstituted HDL (rHDL) preparation reduced ex vivo platelet activation in diabetic subjects, likely by promoting cholesterol efflux from platelets. Human genome wide association studies have revealed novel genetic loci associated with coronary heart disease. One such locus resides in LNK/SH2B3 which in mice is expressed in hematopoietic cells and suppresses thrombopoietin signaling via its receptor MPL. Using human cord blood, we recently showed that the common TT risk genotype (R262W) of LNK is associated with expansion of hematopoietic stem cells and enhanced megakaryopoiesis, demonstrating reduced LNK function and increased MPL signaling. To model the human reduced LNK function associated with the TT risk genotype, we used Lnk^{−/−} mice. In mice, hematopoietic Lnk deficiency led to accelerated arterial thrombosis, but only in the setting of hypercholesterolemia. In platelets LNK deficiency increased MPL signaling, while cholesterol loading decreased SHIP-1 activation, acting convergently to increase AKT and platelet activation. Increased platelet activation combines with hypercholesterolemia induced myelopoiesis to promote pro-thrombotic platelet/leukocyte aggregate formation and accelerated atherogenesis. Atherosclerotic lesions were characterized by larger size, increased necrotic cores and increased leukocyte-derived DNA nets. These findings suggest the possibility that in humans, the common LNK variant rs3184504 impacts platelet activation in a fashion dependent on plasma cholesterol levels. In light of these findings, we propose the following studies as an investigator initiated study. The primary goal is to assess the impact of Evolucumab therapy on platelet function of FH patients.
in a randomized, double blind study. The secondary goal is to determine if platelet activation or the response to Evolucumab therapy is modified by rs3184504 polymorphism. We believe that these investigations will complement ongoing studies to demonstrate that Evolucumab reduces atherothrombotic risk and aid the decision-making as to whether Evolucumab can reduce the atherothrombotic risk in ACS patients.

Lay Abstract:

Cardiovascular disease (CVD) is the leading cause of death worldwide. The majority of CVD are due to atherothrombotic events as a result of advance of atherosclerosis. A causal role of hypercholesterolemia (HC) in atherosclerosis has been well documented. The consensus is that increased plasma LDL cholesterol levels cause excessive LDL cholesterol accumulation in the arterial walls that leads to subsequent pathological changes and causes atherosclerosis. Therapies targeting HC and reducing plasma LDL cholesterol levels as a strategy to reduce the risk of CVD has proven to be a huge success. Platelets are anucleate blood cell components and have a critical role in atherothrombosis, as an important cell component of thrombus in atherothrombosis. There is emerging evidence that HC impacts platelets function, enhances platelet reactivity to various biological stimuli and therefore increases the risk of atherothrombosis. However, the underlying mechanism of this impact of HC on platelet function is not well understood and the implication of this mechanism in prevention or therapies of atherothrombosis has not been tested. In this study, we propose to examine the impact of Evolucumab, an antibody against PCSK9 and causing marked decrease of plasma LDL cholesterol levels in HC human subjects, on platelet functions in HC patients. We hypothesize that Evolucumab reverses the heighted platelet reactivity in HC subjects. Genetic factors demonstrate to be important in inflammatory and metabolic diseases such as CVD. There is evidence that rs3184504, a genetic polymorphism in humans, could impact development of CVD. We have shown in animal models that genetic defect resembling rs3184504 alters arterial thrombosis particularly in HC, likely via a mechanism that alters platelet function and reactivity. Thus, we hypothesize that rs3184504 polymorphism modifies human platelet activation in HC and the response of platelets to Evolucumab therapy. In the proposed studies, we will recruit HC patients and Evolucumab or placebo will be administered. The platelet function as well as plasma levels of certain platelet activation markers will be assessed. We predict that Evolucumab will decrease platelet activation in association with reduced plasma LDL cholesterol levels. We also predict that this effect will be modified by rs3184504 polymorphism.

Abbreviated Submission:
The IRB has an abbreviated submission process for multicenter studies supported by industry or NIH cooperative groups (e.g., ACTG, HVTN, NCI oncology group studies, etc.), and other studies that have a complete stand-alone protocol. The process requires completion of all Rascal fields that provide information regarding local implementation of the study. However, entering study information into all of the relevant Rascal fields is not required, as the Columbia IRBs will rely on the attached stand-alone (e.g., sponsor's) protocol for review of the overall objectives.

If you select the Abbreviated Submission checkbox and a section is not covered by the attached stand-alone protocol, you will need to go back and provide this information in your submission.

Potential Risks:
Provide information regarding all risks to participants that are directly related to participation in this protocol, including any potential for a breach of confidentiality. Risks associated with any of the items described in the
Procedures section of this submission should be outlined here if they are not captured in a stand-alone protocol. Risks of procedures that individuals would be exposed to regardless of whether they choose to participate in this research need not be detailed in this section, unless evaluation of those risks is the focus of this research. When applicable, the likelihood of certain risks should be explained and data on risks that have been encountered in past studies should be provided.

[ ] Abbreviated Submission - This information is included in an attached stand-alone protocol. Proceed to the next question.

The risks of having blood drawn include soreness and bruising at the puncture site, and there may be some discomfort during the blood draw. Rarely, people can feel lightheaded or faint. There is a small risk of infection any time blood is drawn. The amount of blood to be taken is small, and is therefore not expected to have any significant effect. Evolocumab is a humanized IgG2 monoclonal antibody that targets circulating PCSK9 secreted primarily from hepatocytes. Hypersensitivity reactions (e.g. rash, urticaria) have been reported in patients treated with Evolocumab, including some that led to discontinuation of therapy. If signs or symptoms of serious allergic reactions occur, discontinue treatment Evolocumab, treat according to the standard of care, and monitor until signs and symptoms resolve. In the previous published clinical studies such as the study by Sabatine MS et al in New England Journal of Medicine, 2015, 372:1500, the overall adverse events in Evolocumab group were not significantly different from that of the control group. Nevertheless, close monitoring of the potential adverse events will be conducted. These potential adverse events include headache, limb pain, fatigue, arthralgia, nasopharyngitis, upper respiratory tract infection, back pain, nausea, neurocognitive events, elevated plasma levels of alanine or aspartate aminotransferase or creatine kinase. Injection-site reactions will be monitored as well.

Potential Benefits:
Provide information regarding any anticipated benefits of participating in this research. There should be a rational description of why such benefits are expected based on current knowledge. If there is unlikely to be a direct benefit to participants/subjects, describe benefits to society. Please note that elements of participation such as compensation, access to medical care, receiving study results, etc. are not considered benefits of research participation.

[ ] Abbreviated Submission - This information is included in an attached stand-alone protocol. Proceed to the next question.

There is no other direct benefit to participants of this study. There may be future benefit to society in that this research will complement ongoing studies to demonstrate that Evolocumab reduces athero-thrombotic risk and aid the decision-making as to whether Evolocumab can reduce the atherothrombotic risk in acute coronary syndrome patients.

Alternatives:
If this research involves an intervention that presents greater than minimal risk to participants, describe available alternative interventions and provide data to support their efficacy and/or availability. Note, participants always have the option not to participate in research.

[ ] Abbreviated Submission - This information is included in an attached stand-alone protocol. Proceed to the next question.

The alternative is not to participate in this research study. If the patient does not participate in the study, it will not affect the treatment the patient will receive from Dr. Ginsberg. The patients who decide not to participate in the study will be treated according to guidelines and Dr. Ginsberg's experience and knowledge.

Data and Safety Monitoring:
Describe how data and safety will be monitored locally and, if this is a multi-center study, how data and safety...
will be monitored across sites as well.

[ ] Abbreviated Submission - This information is included in an attached stand-alone protocol. Proceed to the next question

For data monitoring, all data and information generated from the study will be stored in encrypted, password protected university or hospital computers. All sensitive data will be stored in Crown and iNYP systems of CUMC. A unique alphanumeric code number will be assigned to each study subject. Patients will be randomized by Research Pharmacy of CUMC and given the testing drug or placebo and the coded data will be securely stored electronically. The coded blood samples will be given to the researcher who will perform the platelet functional assays but will not have access to the data secured by Research Pharmacy of CUMC or any other sensitive data stored in Crown and iNYP systems. Once the study is completed on the entire cohort, all the data and results from different arms involved in the study, including pharmacy, clinic and research labs, will be evaluated and analyzed by the investigators as the coded data. For safety monitoring, we will get baseline blood test for lipids, complete blood count analysis including platelets as well as urinalysis, metabolic panel, hepatic panel. At day 21, day 28 and day 88, we will repeat all the tests. Dr. Henry Ginsberg will review the test results. Any abnormal test will be repeated within 2 weeks. Reductions in hemoglobin levels will lead to measurement of haptoglobin and reticulocyte counts. Based on the results of the tests, Dr. Henry Ginsberg will decide and discuss with the patients about what options are available, including stop of the treatment and withdraw from the study.

### Subjects

Unless otherwise noted, the information entered in this section should reflect the number of subjects enrolled or accrued under the purview of Columbia researchers, whether at Columbia or elsewhere.

**Target enrollment:**
24

**Number anticipated to be enrolled in the next approval period:**
11

**Does this study involve screening/assessment procedures to determine subject eligibility?**
Yes

**Target accrual:**
22

**Number anticipated to be accrued in the next approval period:**
11

**Is this a multi-center study?**
No

**Does this study have one or more components that apply to a subset of the overall study population (e.g. Phase 1/2, sub-studies)?**
No

**Target Enrollment Demographics:**

#### Population Gender

<table>
<thead>
<tr>
<th>Females</th>
<th>Males</th>
<th>Non Specific</th>
</tr>
</thead>
<tbody>
<tr>
<td>0%</td>
<td>0%</td>
<td>100%</td>
</tr>
</tbody>
</table>

#### Population Age

<table>
<thead>
<tr>
<th>Age</th>
<th>Non Specific</th>
</tr>
</thead>
<tbody>
<tr>
<td>0-7</td>
<td>0%</td>
</tr>
<tr>
<td>8-17</td>
<td>0%</td>
</tr>
<tr>
<td>18-65</td>
<td>90%</td>
</tr>
<tr>
<td>&gt;65</td>
<td>10%</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Population Age</th>
<th>Non Specific</th>
</tr>
</thead>
<tbody>
<tr>
<td>0%</td>
<td>100%</td>
</tr>
</tbody>
</table>

IRB-AAAR1041
Population Race

<table>
<thead>
<tr>
<th></th>
<th>American Indian/Alaskan</th>
<th>Asian</th>
<th>Native Hawaiian or Other Pacific Islander</th>
<th>Black or African American</th>
<th>White</th>
<th>More than One Race</th>
<th>Non-Specific</th>
</tr>
</thead>
<tbody>
<tr>
<td>Percentage</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>100%</td>
</tr>
</tbody>
</table>

Population Ethnicity

<table>
<thead>
<tr>
<th></th>
<th>Hispanic or Latino</th>
<th>Not Hispanic or Latino</th>
<th>Non-Specific</th>
</tr>
</thead>
<tbody>
<tr>
<td>Percentage</td>
<td>0%</td>
<td>0%</td>
<td>100%</td>
</tr>
</tbody>
</table>

Vulnerable Populations as per 45 CFR 46:

Will children/minors be enrolled
No

Will pregnant women/fetuses/neonates be targeted for enrollment?
No

Will prisoners be targeted for enrollment?
No

Other Vulnerable Populations:
- [ ] Individuals lacking capacity to provide consent
- [ ] CU/NYPH Employees/Residents/Fellows/Interns/Students
- [x] Economically disadvantaged
- [ ] Educationally disadvantaged
- [ ] Non-English speaking
- [ ] Other Vulnerable populations
- [ ] None of the Populations listed above will be targeted for Enrollment

Subject Population Justification:
This population is selected based on the fact they already have a clinical diagnosis of familial hypercholesterolemia (FH) and are referred to Dr. Ginsberg’s Lipid Practice for treatment with PCSK9 inhibitor, and with LDL cholesterol levels >100 mg/dl on baseline treatment with statins and/or ezitimibe. FH affects 1 in 300 people and all races and ethnicities are affected by FH.

Does this study involve compensation or reimbursement to subjects?
Yes

Describe and justify reimbursement/compensation:
We plan to offer free drug to the participants for the entire study period as compensation for their participation. We also will reimburse for the extra visit travel, $50/visit including public transportation or gas and parking.

Are subjects eligible for compensation of $600 or more in a calendar year?
No

Attached Attestation

<table>
<thead>
<tr>
<th>Type</th>
<th>Principal Investigator</th>
<th>Date Created</th>
</tr>
</thead>
<tbody>
<tr>
<td>C</td>
<td>Nan Wang</td>
<td>11/29/2016</td>
</tr>
</tbody>
</table>
### Attached HIPAA Forms

<table>
<thead>
<tr>
<th>Number</th>
<th>Type</th>
<th>Title</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>AAAN5346</td>
<td>A</td>
<td>Impact of LDL cholesterol lowering by Evolocumab on platelet HIPAA</td>
<td>Approve</td>
</tr>
</tbody>
</table>

### Attached Consent Forms

<table>
<thead>
<tr>
<th>Number</th>
<th>Copied From</th>
<th>Form Type</th>
<th>Title</th>
<th>Active/InActive</th>
<th>Initiator</th>
</tr>
</thead>
<tbody>
<tr>
<td>AAAU4168</td>
<td></td>
<td>Consent</td>
<td>Impact of LDL cholesterol lowering by Evolocumab on platelet activity study consent form</td>
<td>Inactive</td>
<td>Nan Wang (nw30)</td>
</tr>
</tbody>
</table>

### Documents

<table>
<thead>
<tr>
<th>Archived</th>
<th>Document Identifier</th>
<th>Document Type</th>
<th>File Name</th>
<th>Active</th>
<th>Stamped</th>
<th>Date Attached</th>
<th>CreatedBy</th>
</tr>
</thead>
<tbody>
<tr>
<td>No</td>
<td>Consent</td>
<td>Consent Form/Addendum</td>
<td>Consent Form 2-7-17.docx</td>
<td>N</td>
<td>02/07/2017</td>
<td>Nan Wang (nw30)</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>Consent Form 2-7-17</td>
<td>Consent Form/Addendum</td>
<td>Consent Form 2-7-17.pdf</td>
<td>Y</td>
<td>02/09/2017</td>
<td>Deirdre Lombardi (dl2971)</td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>IRB Consent form</td>
<td>Consent Form/Addendum</td>
<td>Human Study Consent Form.docx</td>
<td>Y</td>
<td>01/19/2017</td>
<td>Nan Wang (nw30)</td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>IRB consent form</td>
<td>Consent Form/Addendum</td>
<td>IRB Human Study Consent Form.pdf</td>
<td>Y</td>
<td>11/29/2016</td>
<td>Nan Wang (nw30)</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>Study Design</td>
<td>Other</td>
<td>IRB study design 2-7-17.docx</td>
<td>Y</td>
<td>02/07/2017</td>
<td>Nan Wang (nw30)</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>Response to IRB review</td>
<td>Other</td>
<td>Response to comments.docx</td>
<td>Y</td>
<td>02/07/2017</td>
<td>Nan Wang (nw30)</td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>Response to IRB review</td>
<td>Other</td>
<td>response to IRB review 1-18-17.docx</td>
<td>Y</td>
<td>01/19/2017</td>
<td>Nan Wang (nw30)</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>TC0087 certificate Nan Wang</td>
<td>Other</td>
<td>TC0087 certificate 01 Nan Wang.pdf</td>
<td>Y</td>
<td>01/18/2017</td>
<td>Nan Wang (nw30)</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>TC0087 certificate for Carrie Welch</td>
<td>Other</td>
<td>TC0087 certificate.CWelch.pdf</td>
<td>Y</td>
<td>01/18/2017</td>
<td>Nan Wang (nw30)</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>TC 1450 certificate Wei Wang</td>
<td>Other</td>
<td>TC1450 certificate for Wei.pdf</td>
<td>Y</td>
<td>11/29/2016</td>
<td>Nan Wang (nw30)</td>
<td></td>
</tr>
<tr>
<td>Archived</td>
<td>Document Identifier</td>
<td>Document Type</td>
<td>File Name</td>
<td>Active</td>
<td>Stamped</td>
<td>Date Attached</td>
<td>CreatedBy</td>
</tr>
<tr>
<td>----------</td>
<td>---------------------</td>
<td>---------------</td>
<td>-----------</td>
<td>--------</td>
<td>-----------</td>
<td>---------------</td>
<td>--------------------</td>
</tr>
<tr>
<td>Yes</td>
<td>IRB study design</td>
<td>Standalone/Sponsor's Protocol</td>
<td>IRB study design 1-19-17.docx</td>
<td>Y</td>
<td>01/19/2017</td>
<td></td>
<td>Nan Wang (nw30)</td>
</tr>
</tbody>
</table>