



## **PROTOCOL**

Study Title:  
**Intravenous Toxicity Study in Rats Following a  
Single Administration of rhPDGF-BB**

Study Number: **026691**

Sponsor Study Number: **RnD-120710-01P**

**Testing Facility:**  
Ricerca Biosciences, LLC  
Toxicology and Pharmacology  
7528 Auburn Road  
Concord OH 44077

## ***Introduction***

### **Study Title**

Intravenous Toxicity Study in Rats Following a Single Administration of rhPDGF-BB.

### **Objective(s)**

The objective of this study is to determine the systemic toxicity of recombinant human platelet-derived growth factor-BB (rhPDGF-BB) administered by intravenous injection.

### **Sponsor**

BioMimetic Therapeutics, Inc.  
389 Nichol Mill Lane  
Franklin, TN 37067

Sponsor Representative:

Luis Solchaga, Ph.D.  
Phone: (615) 236-4511  
Cell: (216) 965-4866  
Fax: (615) 236-4470  
email: lsolchaga@biomimetics.com

### **Testing Facility**

Toxicology and Pharmacology  
Ricerca Biosciences, LLC  
7528 Auburn Road  
Concord OH 44077

### **Study Conduct (GLP)**

This study will be performed according to the protocol, testing facility standard operating procedures and the "Good Laboratory Practice" regulations of the United States Food and Drug Administration (21 CFR Part 58).

### **Responsible Personnel**

Study Director:

Eric J. Lubert, Ph.D.  
Phone: (440) 357-3468  
Fax: (440) 357-3800  
email: eric.lubert@ricerca.com

Veterinarian:

Stanley D. Dannemiller, D.V.M., M.S., DACLAM

Dose Formulation Stability\*:

Ken Shockley, Ph.D.  
Director of Laboratory Operations  
BioMimetic Therapeutics, Inc.  
389 Nichol Mill Lane  
Franklin, TN 37067

Ophthalmology\*:

staff veterinarian

Clinical Pathologist\*:

James R. Szabo, D.V.M., Ph.D., DACVP

Pathologist\*:

James R. Szabo, D.V.M., Ph.D., DACVP or  
Karen Regan, D.V.M., DABT, DACVP

*\* contributing report to be included in the final report*

## Proposed Schedule of Events

First Day of Dosing:	7-December-2010
In-Life Termination:	20-December-2010

## *Materials and Methods*

### Test Article

#### Identification

The Sponsor's designation of the test article is rhPDGF-BB.

#### Composition and Purity

The test article will be supplied by the Sponsor as pre-formulated solutions. The composition and purity of the test article are the responsibility of the Sponsor. A Certificate of Analysis or other appropriate documentation for each lot of the test article to be used and each formulation level will be provided by the Sponsor prior to finalization of the final report. A copy of all appropriate documentation will be included in the final report. The test article will be used without correction for purity or salt content.

#### Storage Conditions

The test article formulations will be stored in their original container(s) in a refrigerator at 2-8°C.

#### Stability

The stability of the test article formulations, under the storage conditions to be used in this study, is the responsibility of the Sponsor. Following the completion of dosing, all remaining test article formulations will be shipped back to the Sponsor for stability assessment.

#### Retention Sample

The Sponsor will maintain a retention sample of each lot of test article under the refrigerated conditions of bulk test article storage in accordance with applicable regulations and guidelines for pre-clinical studies.

#### Test Article Accountability and Disposition

Test article formulation accountability will be maintained according to testing facility SOPs. Following the completion of dosing, the remaining test article formulations will be shipped back to the Sponsor.

### Vehicle

The vehicle will be 20 mM Sodium Acetate (NaOAc), pH 6.0±0.5. All pertinent information will be retained in the study file (description, lot number, etc.).

### Preparation, Storage, and Analysis of Dosing Formulations

The Sponsor will prepare and provide Ricerca with an appropriate amount of the 0.3 mg/mL and 3.00 mg/mL dosing formulations. The formulations will be stored in a refrigerator at 2-8°C. On the day of dosing, a portion of the 0.3 mg/mL solution will be diluted 1:10 in vehicle

to create the 0.03 mg/mL solution. Ricerca will provide three 1 mL samples from the 0.03 mg/mL solution for homogeneity and concentration analysis by the Sponsor.

## Safety Precautions

Standard laboratory safety procedures will be employed for handling the dose formulations. Specifically, gloves and eye protection will be worn while administering doses. The manufacturer(s) of the test article will supply a Material Safety Data Sheet (MSDS) and/or other pertinent documentation regarding safety.

## Test System (Animals and Animal Care)

### Species, Source and History

CrI:CD(SD) Sprague-Dawley rats will be obtained from Charles River Laboratories. The animals will be laboratory bred and experimentally naïve. The females will be nulliparous and nonpregnant. Rats were chosen because they are a species that is commonly used for nonclinical toxicity evaluations.

### Number, Sex, Age, and Body Weight Range

176 rats (88 male and 88 female) will be ordered. From these animals, 80 males and 80 females will be used. Animals will be as uniform in age as possible. The rats will be prepubertal to young adult, approximately 7-9 weeks of age and approximately 150-400 g at the time of randomization.

The dates of birth for the animals will be retained in the study records, and the age and body weight ranges will be specified in the report.

### Identification

Each animal will be identified by a unique animal number via an ear tag and matching cage card.

### Housing and Sanitation

The animals will be housed individually in stainless steel, wire-bottom cages. Primary enclosures will be as described in *Guide for the Care and Use of Laboratory Animals* (National Academy Press, Washington DC, 1996). All animals will be kept in one room. No additional studies or other species will be housed in the room. Animal room and cage cleaning will be performed according to testing facility SOPs.

### Environmental Conditions

Temperature:	64 - 79°F (18 - 26°C)
Humidity:	30 - 70%
Light:	An alternating approximate 12-hour light/dark cycle will be maintained, except during study-specific procedures. Addition or loss of time due to Daylight Savings time adjustments will not be considered as deviations.
Ventilation:	The airflow will be at least 10 air changes per hour with 100% fresh air (no air recirculation).

The environmental conditions of the animal room are maintained per *Guide for the Care and Use of Laboratory Animals* (National Academy Press, Washington DC, 1996) and will be continuously monitored and documented. Transient fluctuations outside this range may occur. Transient environmental fluctuations that persist for less than 2 hours, provided that

temperatures do not exceed 85 °F, will be considered to have no adverse impact on the study and will not be listed in the final report.

### **Feed and Water**

Harlan Teklad Certified Global Diet<sup>®</sup> 2016C will be available *ad libitum* except during protocol-specified periods of fasting. No contaminants are known to be present in the certified diet that would interfere with the results of the study. Analysis of diet will be limited to that performed by the manufacturer.

Tap water will be available *ad libitum*. Water is supplied by the Painesville Water District and is routinely analyzed to monitor chemical and bacterial levels and to determine priority pollutants.

Results of the food and water analyses are reviewed periodically by a staff veterinarian or designee. All food and water analysis records will be archived at Ricerca Biosciences.

### **Acclimation, Pre-study Health Screen, and Selection Criteria**

All animals will be acclimated to laboratory conditions for a minimum of 5 days prior to the start of dosing. During that period, the health status of the animals will be evaluated in accordance with accepted veterinary practice. Only animals considered acceptable based on health status will be assigned to study.

### **Humane Care of Animals**

Treatment of the animals will be in accordance with the conditions specified in *Guide for the Care and Use of Laboratory Animals* (National Academy Press, Washington DC, 1996).

### **Assignment to Study Groups, Control of Bias**

Prior to assignment, for a given sex, no animal will be considered for assignment if out of the  $\pm 20\%$  range from their corresponding group mean body weight. Animals judged acceptable for use on the study will be randomly assigned to study groups using a computer-generated process of randomization based on body weight to control bias. The mean body weight for each group will differ by no more than 5% from the control group (Group 1) at the time of group assignment. Animals not used on the study will be removed from the animal room the day following initial dosing of all animals.

### **Study Design**

Animals will receive rhPDGF-BB (test article) or vehicle control (20 mM NaOAc, pH 6.0 $\pm$ 0.5) via a single intravenous injection and terminated in accordance with the experimental design presented in the tables below. The day of dosing will be designated as Day 1 of the study.

### Group Assignment and Dose Levels

Dose Group	#Animals (M/F)	Test Article	Dose Level (mg/kg)	Dose Conc. (mg/mL)	Dose Volume (mL/kg)	Number of Animals for Necropsy (M/F)	
						Day 2	Day 14
1	20/20	NaOAc (Vehicle)	0.000	0.00	1.4	10/10	10/10
2	20/20	rhPDGF-BB	0.042	0.03	1.4	10/10	10/10
3	20/20	rhPDGF-BB	0.420	0.30	1.4	10/10	10/10
4	20/20	rhPDGF-BB	4.200	3.00	1.4	10/10	10/10

The animals will be evaluated for changes in clinical signs, body weight, and other parameters as described below.

### Route/Dose

The intravenous route of administration represents the highest systemic exposure levels to the test article. The dose levels were selected based on a maximum clinical dose of 41.4 µg/kg based on a 70 kg patient.

The test article will be administered by slow bolus intravenous injection into the tail vein at a dose volume of approximately 1.4 mL/kg body weight. Control animals will receive vehicle only at the same dose volume.

## In-Life Observations and Measurements

### Observations

Animals will be observed for viability at least once in the morning and once in the afternoon, at least 4 hours apart, throughout the study.

Cageside observations will be made and recorded at 1, 2.5, and 4 hours postdose. Thereafter, cageside observations will be made and recorded once daily at approximately the same time each day.

A detailed clinical examination will be performed on all animals prior to the initiation of dosing and then weekly during the study.

Animals appearing ill, and/or in pain or distress, will be brought to the attention of the Study Director and a staff veterinarian and may be euthanized in accordance with the Testing Facility SOPs.

### Unscheduled Observations

Findings observed at times other than scheduled observation times will be recorded on an unscheduled observation form and/or documented in Provantis, an electronic database.

### Body Weights

Prior to start of dosing, body weights will be measured at least twice including at randomization on Day -1. During the treatment and nondosing periods, body weights will be measured once weekly, at the end of the study week. In addition, fasted body weights will be measured prior to necropsy for calculation of organ/body weight ratios.

### Food Consumption

Food consumption data will be collected for all animals prior to randomization. Following randomization on Study Day -1, animals scheduled for necropsy on Study Day 14 will have

food consumption data collected weekly. **NOTE:** Animals scheduled for necropsy on Study Day 2 will not have food consumption data collected following randomization. At the discretion of the Study Director, the pretest food consumption data may be collected for less than 7 days.

### Ophthalmic Examination

An ophthalmic examination will be conducted on all animals prior to dosing prior to their scheduled necropsy on either Study Day 2 or Study Day 14 (~2 weeks post dose). Indirect ophthalmoscopy will be performed by a staff veterinarian to examine the fundus, vitreous, and anterior chamber. Prior to the examination, an ophthalmic mydriatic solution (typically 1% tropicamide) will be instilled into each eye to facilitate the examination.

### Clinical Pathology Evaluation

A whole blood sample will be collected from any extra animals on Study Day 2. Animals will be bled from the retro-orbital plexus under CO<sub>2</sub>/O<sub>2</sub> anesthesia. Animals will be fasted overnight prior to this blood collection. The sample will be allowed to clot prior to centrifugation. Resultant serum samples will be stored frozen at approximately -70°C until shipped to the Sponsor. **NOTE:** These serum samples will be retained by the Sponsor in the event that any future testing is requested. Should any testing be conducted by the Sponsor or their designee, the results will not be included in the final report or any amendments to the final report for this study.

Whole blood samples will be collected from all Group 1-4 animals prior to their scheduled necropsy on either Study Day 2 or Study Day 14 (~2 weeks post dose) for the evaluation of hematology, coagulation, and serum chemistry parameters. Animals will be bled from the retro-orbital plexus under CO<sub>2</sub>/O<sub>2</sub> anesthesia. Animals will be fasted overnight prior to this blood collection. Animals will not undergo necropsy until after the blood has been collected and the samples judged to be acceptable by the clinical pathology group.

### Hematology

An appropriate amount of blood will be collected into EDTA-containing tubes. The whole blood samples will be analyzed for the following parameters.

Red blood cells (RBC) (count and morphology)	Mean corpuscular volume (MCV)
Hematocrit (HCT)	Mean corpuscular hemoglobin (MCH)
Hemoglobin concentration (HGB)	Mean corpuscular hemoglobin concentration (MCHC)
Platelet count (PLAT)	Mean platelet volume (MPV)
White blood cells (WBC) (total and differential)	Absolute reticulocyte count (ABSRET)

### Coagulation

An appropriate amount of blood will be collected in tubes containing sodium citrate and then centrifuged to obtain plasma for the determination of prothrombin time (PT), and activated partial thromboplastin time (APTT).

### Serum Chemistry

An appropriate amount of blood will be collected in tubes without anticoagulant. The sample will be allowed to clot and then will be centrifuged to obtain serum. The serum will be analyzed for the following parameters. Any remaining serum will be stored frozen at

approximately -70°C until shipped to the Sponsor. **NOTE:** These serum samples will be retained by the Sponsor in the event that any future testing is requested. Should any testing be conducted by the Sponsor, or their designee, the results will not be included in the final report or any amendments to the final report for this study.

Sodium (NA)	Calcium (CA)
Potassium (K)	Inorganic phosphorus (PHOS)
Chloride (CL)	Glucose (GLU)
Total bilirubin (TBILI)	Urea nitrogen (BUN)
Alkaline phosphatase (ALKP)	Total protein (TPRO)
Lactate dehydrogenase (LDH)	Albumin (ALB)
Aspartate aminotransferase (AST)	Globulin (GLOB)
Alanine aminotransferase (ALT)	Albumin/globulin ratio (A/G)
Gamma-glutamyltransferase (GGT)	Cholesterol (CHOL)
Creatine phosphokinase (CK)	Triglycerides (TRIG)
Creatinine (CREA)	

All serum samples to be retained by the Sponsor (i.e., naïve extra and prior to necropsy) will be shipped on dry ice to Attn: Luis Solchaga, BioMimetic Therapeutics, Inc., 389 Nichol Mill Lane, Franklin, TN 37067.

## Urinalysis

Urine will be collected overnight from all study animals prior to their scheduled termination. The following urinalysis parameters and their collection times will be recorded and evaluated for each animal while housed individually.

Bilirubin	Nitrite
Clarity	Occult blood
Color	pH
Glucose	Protein
Ketones	Specific gravity
Leukocytes	Urobilinogen
Microscopic examination of sediment	Volume

## Termination

### Unscheduled Termination

If an animal dies or is terminated moribund during the study, it will be subjected to a necropsy as soon as possible. If a necropsy cannot be performed within 30 minutes of the observation, the abdominal cavity will be opened and the carcass will be refrigerated (not frozen) to minimize tissue autolysis. Duration of refrigeration will be recorded.

All animals considered moribund will be euthanized on the day that determination is made. Animals considered moribund will be terminated by exsanguination (severing the abdominal aorta while under deep anesthesia induced with CO<sub>2</sub>/O<sub>2</sub>).

### Scheduled Termination

All surviving study animals will be euthanized and subjected to a necropsy following the protocol-prescribed number of days. Animals will be terminated by exsanguination (severing the abdominal aorta while under deep anesthesia induced with CO<sub>2</sub>/O<sub>2</sub>).

### Necropsy

A necropsy with tissue collection will be conducted on all study animals found dead or terminated following dosing on Day 1. The necropsy will include examination of:

- Carcass and muscular/skeletal system
- All external surfaces and orifices
- Cranial cavity and external surface of the brain
- Neck with associated organs and tissues
- Thoracic, abdominal, and pelvic cavities with their associated organs and tissues

All abnormalities will be described and recorded.

### Organ Weights

For all animals euthanized at scheduled necropsies, the designated organs listed in the tissue collection table will be weighed. Paired organs will be weighed together. The pituitary and thyroids/parathyroids will be weighed post-fixation. Organ weights will not be taken from animals found dead or euthanized moribund.

Organ/body weight ratios will be calculated using the terminal fasted body weight obtained prior to necropsy. Organ/brain weight ratios will also be calculated.

### Tissue Preservation

The following tissues and organs will be collected from all animals and will be preserved in 10% neutral-buffered formalin with the exception of the testes, epididymides, and eyes. Testes, epididymides, and eyes with optic nerve attached will be fixed in Modified Davidson's Solution for ~24 - 48 hours, rinsed with water, and then transferred to 10% neutral buffered formalin for storage.

**Tissue Collection Table**

Tissue	Submitted at Necropsy	Organ Weight	Histopathology
Animal ID	X		
Adrenal gland (2)	X	X	X
Aorta	X		X
Bone (femur)	X		X
Bone (sternum)	X		X
Bone marrow (femur)	X		X
Bone marrow (sternum)	X		X
Brain	X	X	X
Cervix <sup>a</sup>	X	X <sup>a</sup>	X
Epididymides (2)	X		X
Esophagus	X		X
Eyes (2)	X		X

Tissue	Submitted at Necropsy	Organ Weight	Histopathology
Gross lesions	X		X
Heart	X	X	X
Injection site(s)	X		X
Intestine, cecum	X		X
Intestine, colon	X		X
Intestine, duodenum	X		X
Intestine, jejunum	X		X
Intestine, ileum	X		X
Intestine, rectum	X		X
Kidneys (2)	X	X	X
Liver	X	X	X
Lungs	X	X	X
Lymph node, cervical	X		X
Lymph node, mesenteric	X		X
Mammary gland (both sexes)	X		X
Nerve, optic	X		X
Nerve, sciatic	X		X
Ovaries (2)	X	X	X
Pancreas	X		X
Parathyroid glands (2) <sup>b</sup>	X	X <sup>b</sup>	X
Pituitary	X	X	X
Prostate	X		X
Salivary gland (mandibular)(2)	X		X
Seminal vesicles	X		X
Skeletal muscle ( <i>biceps femoris</i> )	X		X
Skin	X		X
Spinal cord, cervical	X		X
Spinal cord, thoracic	X		X
Spinal cord, lumbar	X		X
Spleen	X	X	X
Stomach	X		X
Testes (2)	X	X	X
Thymus	X	X	X
Thyroid gland (2) <sup>b</sup>	X	X <sup>b</sup>	X
Tongue	X		X
Trachea	X		X
Urinary bladder	X		X
Uterus <sup>a</sup>	X	X <sup>a</sup>	X

Tissue	Submitted at Necropsy	Organ Weight	Histopathology
Vagina	X		X

<sup>a</sup> Uterus weighed with cervix attached.

<sup>b</sup> Thyroid weighed with parathyroids attached.

### Histopathology (Groups 1 to 4)

All collected tissues will be shipped and processed to slides at Vet Path Services Inc., 6450 Castle Dr., Mason, OH 45040. All tissues listed above will be examined by a trained veterinary pathologist at Ricerca Biosciences (or their designee).

### Data Collection

Animal data, such as observations, body weights, clinical pathology, necropsy, and organ weights, will be collected and reported electronically using Provantis™, Version 8 (Instem LSS Ltd. Staffordshire, UK). Provantis was validated at Ricerca Biosciences. Data not collected by Provantis by design or due to technical problems will be hand-recorded on paper or collected electronically by other means. This data may be transferred to and reported from Provantis. Tables generated from Provantis will be exported as Microsoft Word documents and verified to assure accuracy.

### Statistical Analysis

For comparative statistics, data will be evaluated using the Levene Test for homogeneity of variances and the Shapiro-Wilks Test for normality of distributions, with significance at  $p \leq 0.05$ . Data determined to be homogeneous and of normal distribution will be evaluated for analysis of variance (ANOVA). If the ANOVA verifies significance at  $p \leq 0.05$ , pairwise comparisons of each treated group with the control group will be made using a parametric test, e.g., a Dunnett Test, to identify statistical differences ( $p \leq 0.05$ ). Data determined to be nonhomogeneous or of nonnormal distribution will be evaluated using a Kruskal-Wallis Test for group factor significance. If significance ( $p \leq 0.05$ ) exists between groups, a nonparametric test, e.g., Wilcoxon with Bonferroni-Holm or Dunnett Test (nonparametric) will be used to compare treatment groups to the control group.

Comparative statistics of food consumption data may be limited to the Dunnett Test (parametric) and may exclude pretest food consumption data. Food consumption data from animals where spillage occurs will be excluded from the applicable time period.

At the discretion of the Study Director and following a consultation with the Sponsor, alternative statistics may be conducted or statistical evaluation may be eliminated if there is lack of sufficient data due to excess animal mortality or other pertinent factors.

## Records and Reports

### Records

Records to be maintained will include, but not be limited to, those records necessary to satisfy the reporting requirements listed in the next section. The following will be collected and retained as part of the study file in the archives:

- Animal procurement records including existing prior health records
- Animal health data
- Physical examinations and other pre-study health screen data
- Test article and formulation records

- Dosing data
- Nature, onset, severity, and duration of all gross or visible toxic or pharmacological effects
- Mortality data, if any
- Food consumption data
- Body weight data
- Ophthalmology exam data
- Clinical pathology (hematology, coagulation, serum chemistry, and urinalysis) data
- Necropsy observations
- Organ weight data
- Tissues, blocks, and slides
- Statistical analyses
- Histopathology findings
- Letters or any written communication concerning the study
- Protocol, amendments, and deviations
- Final report

All records and tissue specimens will be retained and archived in compliance with all applicable regulations. Tissues, blocks, and slides will be maintained at VPS Archives, LLC, 6450 Castle Dr., Mason OH 45040. With the exception of data generated for the test article formulation characterization and stability analyses, all other materials, including the raw data and final report, will be archived at Ricerca Biosciences, LLC. Archival of any data generated for the test article formulation characterization and stability analyses will be the responsibility of the Sponsor and/or their designee. After a period of 2 years, the Sponsor will be contacted for disposition instructions for the archival material.

## Reporting

The Study Director will prepare a final report of the results. The report will include all items in the protocol as well as a comprehensive presentation of all data collected in the study. Deviations, if any, to the protocol will be included in the report. The name and address of the facility conducting the study, and dates on which the study was initiated and completed will be included in the report.

## ***Regulatory Compliance***

### **Protocol Changes and Deviations**

Changes to the approved protocol will be in the form of amendments approved by the Study Director and the Sponsor Representative. Amendments will clearly describe the protocol changes and will include the effective date of the change and justification for the change. The Study Director and Sponsor Representative may authorize protocol changes by telephone, facsimile machine or other electronic means if he/she is not physically present at the time urgent or critical changes are required. Any authorization for such changes made as above, must be appropriately documented and followed by a properly prepared written protocol amendment. The amendment must be signed and dated by the Study Director as soon as possible. Protocol deviations will be noted in the study records and reported.

### **Study Audits**

#### **Quality Assurance**

The Testing Facility Quality Assurance Unit will make periodic inspections and examine the data to determine that it is complete, consistent, and well documented. The unit will also examine the final report to assure that it accurately represents the raw data collected in the study.

The Test Site Quality Assurance Unit will be responsible for inspections and audits in accordance with their standard operating procedures. A Quality Assurance Statement listing the phases inspected, dates of inspection, and dates that findings were reported to the Principal Investigator (PI), PI management, Study Director, and Testing Facility Management will be prepared and included in the contributing scientist report.

#### **Sponsor Audits**

The Sponsor may send representatives to inspect the study and the premises during normal working hours during the course of the study and/or to audit the raw data and the final report.

### **Institutional Animal Care and Use Committee**

The requirements for, and use of animals in, this research are the responsibility of the Sponsor, in that the research does not unnecessarily duplicate previous animal experiments and is being conducted in the absence of acceptable nonanimal alternatives to accomplish its objectives. The protocol and any amendments or procedures involving the care or use of animals in this study will be reviewed and approved by Ricerca's Institutional Animal Care and Use Committee (IACUC) prior to the initiation of such procedures (i.e., prior to the start of the study for most protocol-specified procedures).

### **Ownership of the Study**

The Sponsor owns the study; this includes the data, results, raw material, any samples, etc.

## ***Protocol Review***

### ***Quality Assurance***

\_\_\_\_\_  
***Michael J. Keherly, Ph.D.***  
***Ricerca Biosciences, LLC***

***Date:*** \_\_\_\_\_

## ***Approval for Study Initiation***

### ***Sponsor Representative***

\_\_\_\_\_  
***Luis Solchaga, Ph.D.***  
***BioMimetic Therapeutics, Inc.***

***Date:*** \_\_\_\_\_

### ***Study Director***

\_\_\_\_\_  
***Eric J. Lubert, Ph.D.***  
***Scientist, Drug Safety***  
***Ricerca Biosciences, LLC***

***Date:*** \_\_\_\_\_

### ***Testing Facility Management***

\_\_\_\_\_  
***Douglas K. Fuhrer, Ph.D., DABT***  
***Senior Director, Drug Safety***  
***Ricerca Biosciences, LLC***

***Date:*** \_\_\_\_\_