COURSE SYLLABUS

BIOL827: Biological Statistics

Department of Biology, University of Nebraska-Kearney

Term: Fall 2022

Course Credits: 3

Course Delivery: Online - Asynchronous

Instructor

Dr. Jayne Jonas, UNK Department of Biology, jonasj@unk.edu, office: 308.865.8224

Zoom office hours

Mondays and Wednesdays, 11:00AM – 12:00PM Central Time, or by appointment.

Please contact me any time with questions or concerns. If emailing outside of Canvas, please include BIOL827 in the subject line to ensure a timely response. I respond to messages as quickly as I can, generally within 24-48 hours.

Course Overview

This class is divided into two main areas. The first is biological statistics: the collection and analysis of scientific data. The second area is experimental design: how an experimental hypothesis is built and what are the pieces and procedures needed to conduct a successful experiment. The class is not mathematically intensive and relies on the power of computers beyond a few examples done by hand. The class includes both parametric and non-parametric statistics with continuous and categorical variables. Per the UNK Graduate Course Catalog.

Course Learning Objectives

At the completion of the course, you will be able to

- 1. Identify best practices for data management, including creation of metadata, to ensure longevity of datasets.
- 2. Construct testable hypotheses about biological systems and identify appropriate experimental designs to statistically test those hypotheses.
- 3. Calculate descriptive statistics to examine the character of a dataset and evaluate statistical power.
- 4. Conduct and interpret results of standard statistical tests as appropriate for the experimental design used and data collected.
- 5. Apply statistical concepts in critically evaluating research conducted by others.
- 6. Communicate research results that are accurately, concisely, and straightforwardly supported by statistics.

Important University Dates

- First day of the session: August 22, 2022
- University Breaks/Holidays: September 5, 2022, October 17-18, November 23-27
- Last day to withdraw: November 11, 2022
- Last day of the session: December 15, 2022
- See the <u>UNK Academic Calendar</u> for other important University dates

Required Texts and Materials

- Whitlock, M.C., Schluter, D. The Analysis of Biological Data. Freeman-Macmillan, New York.
 - o 2015 Second edition (preferred), or 2020 Third edition
 - To purchase the textbook, visit the course page of the UNK online book supplier: http://unk.textbookx.com/institutional/index.php?action=browse#books/3421169/
- Other:
 - Some required and optional reading materials may be provided via the Library Reserves or require accessing via Library citation finder.
 - Any other course readings, website, and interactives (required or optional) will be posted for download or linked on the Learning Content pages in Canvas.

Required Technology

- As an online course, access to an up-to-date computer and <u>Canvas</u> several times a week is required (checking in daily is encouraged). Use of an obsolete computer operating system may hinder functionality of both Canvas and RStudio.
- All Canvas quizzes should be completed from a computer browser. Issues may arise if using the Canvas mobile app or browser on a mobile device to complete quizzes.
- Application of concepts via statistical software is central to both current research practice and critical evaluation of scientific literature. Familiarity with statistical computing is also seen as a valuable skill by many employers in the biological sciences. In this course, we will use the R program implemented using the free software RStudio Desktop. Both are available for Windows, Mac, and Linux operating systems. Please install both software programs prior to beginning Module 1. No prior knowledge of or experience with this software is needed at the beginning of the course we will start from square one and work on building skills incrementally during the semester. Although current builds of RStudio are designed for 64-bit computers, you can download and install an older build of RStudio compatible with 32-bit computers. For issues with R/RStudio, please contact the instructor.
 - If you prefer using a different program (ex., MSExcel, SAS, SPSS), that is okay as long as it produces comparable output and results as RStudio; I will not provide support for other software.
- You may need to use a word processing software, such as Microsoft Word, Apple Pages, or Google Docs.
 - Word processing programs include tools for typing equations; you should familiarize yourself with the equation editor in the word processing software of your choice. The

equation editor is launched by selecting 'Equation' from the 'Insert' menu in all three word-processing programs listed above.

- Microsoft 365 software is available to UNK students for free download.
- Familiarity with spreadsheet programs such as Microsoft Excel or Google Sheets is helpful.
- Ability to use search engines (for example, <u>Google Scholar</u> or <u>the Calvin Library search engine</u>) to locate scholarly works is expected of graduate students. If you are not already comfortable with this, I encourage you to reach out to me or the <u>Natural Sciences Librarian</u> at Calvin Library for assistance.
- For issues with Canvas or other technologies associated with your university account, please contact the University technology help desk.

Course Organization

There are 16 content modules assigned over 15 weeks in this course. Each module consists of module learning objectives, a set of mini-lectures, assigned readings, and any other assigned learning materials. One or two modules will be covered each week (Monday 12:01 am to the next Monday 11:59 pm; modules overlap on Mondays). All materials will be listed, posted, or linked in Canvas. As a 3-credit course completed in 12 instructional weeks (plus Finals Week), it is expected that you will spend at least 9 - 12 hours per week on this course.

Participation

The more students are engaged in a course, the more they tend to get out of it. I encourage you to spend at least a little time each day with course material or activities though I recognize this is not always possible.

Communication

Please check course announcements in Canvas frequently. This the primary way I will communicate. Please feel free to reach out to me at any time via Canvas messages, email, or office phone. I will respond as quickly as possible, usually within 24-48 hours. If sending an email or leaving a voicemail, please be sure to include BIOL827 in the subject (email) or message (voicemail) so I can prioritize responding to it.

Course Assessments

All work is assigned individually to all students and due by 11:59PM Central Time on the date listed in Canvas and on the course schedule, unless otherwise indicated. If there is a discrepancy between the course schedule and Canvas, the date in the course schedule takes precedence.

Discussion Weekly discussions provide students an opportunity to critically examine a peer-reviewed scientific study and discuss how concepts presented that week apply to it. Importantly, they also allow students to support one another in learning. Students are expected to uphold UNK Values and any other principles of community identified by your group to establish the discussion board as a supportive and inclusive learning space.

Discussion participation: You are expected to compose at least **three posts** for each discussion (more are encouraged!): one in direct response to a discussion prompt on or

before **Thursday** and a response to two other students on or before **Sunday** of the assigned week. The grading rubric and information regarding expectations for discussion participation are available in Canvas. Participation in each discussion is worth 10 points. There will be 15 graded discussions, the lowest discussion score will be dropped (i.e., 14 will count toward the semester grade).

I will follow discussions throughout each module. However, being cognizant that students will be posting at different times during the week and to avoid steering discussion too much, I will generally limit my contributions unless there are areas in need of immediate attention. Following each module, a summary of group discussions will be posted by the discussion leader (see below) to the corresponding Discussion Recap page for review by all students in the course.

Discussion leading: Each student will be assigned **one module to lead discussion** (40 points). Student leaders are also expected to participate in and will receive a separate participation grade for the discussion they are assigned to lead. See the Discussion Leading Assignment in Canvas for complete instructions, schedule, and grading rubric.

Discussion leaders are expected to find a peer-reviewed scientific study (journal article, book chapter, etc.) from any field of biology relevant to that week's topic for the group to critique. The leader wll pose two to three thought-provoking questions about the study and how it relates to the week's topic for their group to discuss.

Materials are due to be posted to the discussion board by the discussion leader no later than 11:59PM Central Time on **Tuesday of the assigned module**. At the conclusion of the module, the leader will write a *brief* summary highlighting the main points discussed for each question and post it to the Discussion Recap page of the corresponding module by the 11:59PM on **the Monday of the next module**.

Problem sets

Each week will have a 15-point problem set assigned except during weeks with an exam. Problem sets will provide students practice applying concepts relevant to each module, handling data, conducting analyses in R, and interpreting output produced by R. Each will be due by 11:59PM on **the Monday of the next module**. There will be a total of 13 graded problem sets, with the lowest score being dropped (i.e., 12 scores will contribute to the semester grade).

A file with detailed instructions and any datafiles will be provided for each problem set in Canvas. Students are expected to read and follow these instructions. Students may work through problem sets collaboratively, but each student must submit their own unique work and will be responsible for that material on quizzes and exams.

Most problem sets will be submitted by entering responses to the questions in the instructions file using a Canvas quiz page. If there is a discrepancy between the instructions document and the Canvas quiz form, the instructions document takes precedence. When entering problem set responses into a Canvas quiz form, students will

not have a time limit in which to complete the assignment or be limited in the number of attempts (the last submission will be graded) until the due date/time.

Quizzes

There will be a 15-point quiz each week related to the learning objectives of the module(s) covered except in weeks with an exam. Quizzes are to be completed by each student independently (i.e., no collaboration with others). They may require students to refer to tables in the text or other materials provided in the module and may include simple calculations requiring a calculator or spreadsheet.

Weekly quizzes must be completed by 11:59PM each **Saturday**. Quizzes are open book/note (no proctor needed), students will not be able to stop then resume once started. Once started, students will have 20 minutes to complete the quiz. Students will have two attempts to take each quiz (highest score kept). Of the 14 weekly quizzes, the lowest two scores will be dropped (i.e., 12 scores will contribute to the semester grade).

Important note: Canvas quizzes should be completed from a computer browser. Issues tend to arise when using the Canvas mobile app or browser on a mobile device.

Exams

There are two unit exams (90 points each) and a comprehensive final exam (180 points). Exams have the following components:

- 1) timed quiz,
- 2) problem set, and
- 3) [final exam only] paper critique.

All exam components are to be completed by each student independently (i.e., no collaboration). All timed quiz portions are open note/open book (proctor not required). Students have one attempt to take the timed exam and will not be able to stop then resume once started. Specific instructions and expectations for exam problem sets (all exams) and critique (final only) will be provided in Canvas one-week prior to being due. Additional office hours may be offered during exam weeks, watch Canvas Announcements.

Unit exams (Modules 5 and 11): Unit exams will cover material relevant to learning objectives of each module in the unit, including the module in which the exam occurs. Exams will be cumulative only in so far as the material in the course builds upon itself. The timed exam will be due by 11:59PM **Saturday of exam week**, and the problem set will be due by 11:59PM **Monday** after the timed exam is due. Once started, students will have 75 minutes to complete the timed portion of the unit exam. Problem sets can be submitted at any time prior to being due.

Comprehensive final exam (Module 17): All portions of the final exam are due in the last week of the semester. The paper critique will be due by 11:59PM on Tuesday. The timed quiz portion of the comprehensive final will be available only on Wednesday (12:01AM to 11:59PM) and must be submitted by 11:59PM that day. Once started, students will have 90 minutes to complete the timed portion of the final exam; otherwise the timed portion of

the final will run similarly to unit exams. The take-home problem set will be due by **5:00PM** on **Thursday of Final's week**.

Basis for final grade

Assessments and point distribution*	Points	% of grade
Module discussions	180	20%
Participation (15 @ 10 points each, 1 dropped)	140	16%
Leading (1 @ 40 points each)	40	4%
Weekly problem sets (13 @ 15 points each, 1 dropped)	180	20%
Quizzes/Exams	540	60%
Quizzes (14 @ 15 points each, 2 dropped)	180	20%
Unit exams (2 @ 90 points each)	180	20%
Final exam	180	20%
Total	900	100 %

^{*}Adjustments may be made if deemed necessary by the instructor

Final letter grades will be assigned following a straight letter scheme (i.e., no +/- except as described under Grading Policy below) as follows:

A: 90 - 100% D: 60 - < 70% B: 80 - < 90% F: < 60%

C: 70 - < 80%

Course Policies

I take my role as your instructor very seriously; I care about how well you do in this course and that you have a challenging and rewarding experience. It is my commitment to you to respond individually to the work you submit in this class and to return your work promptly.

Discussions, problem sets, and weekly quizzes will be returned within one week. Exams will be returned within ten days. If grading will take longer than the times listed here, I will keep you informed of my progress and return your work as soon as I can.

Grading Policy

If you think there was a grading error or do not understand the feedback you receive on graded work, please contact me as soon as possible. If you would like me to regrade your work, requests should be made within three days after the graded work has been returned to you. Regrade requests may result in a lower grade.

Accommodations for cases in which an end of semester grade percentage falls within 0.50% of the next highest letter grade must be requested by the student by 8am on the Tuesday after finals

week. I will take participation and engagement throughout the semester into consideration in deciding whether to make a final grade accommodation. When granted, course letter grade accommodations will result in a half-letter increase (for example, instead of a B an accommodated student would receive an A-). Accommodations will not be considered for semester grade percentages more than 0.50% from the next letter grade.

Late Work Policy

As a student enrolled in this course, one of your responsibilities is to submit course work on time. With that said, I recognize there may be times when you are unable to complete or submit these tasks by their due dates. To accommodate this, one discussion participation, one weekly problem set, and two weekly quiz scores will be dropped. Canvas automatically adjusts your course grade throughout the semester to reflect dropping these values.

Quizzes, Exams: These assessments should be completed within the designated timeframe. Quizzes and exams will not be accepted late unless prior arrangements have been made due to *documented* professional or extenuating personal circumstances (e.g., family emergency, participation in University-sanctioned activities, religious observation, etc.). Please contact me as soon as possible to discuss alternative arrangements.

Discussions, Problem Sets: These assignments will receive a one letter grade deduction (10% of points possible on assignment) for each day late unless prior arrangements have been made or if there are professional or extenuating circumstances (e.g., family emergency, participation in University-sanctioned activities, religious observation, etc.). Late assignments will be accepted up to 4 days late. If more than 4 days late, the assignment will not be accepted and a grade of 0 (zero) will be recorded for that assignment. Please contact me as soon as possible.

Extra Credit Policy

Extra credit opportunities may be provided at the discretion of the instructor.

Final Exam Policy

Final examination week is part of the regular semester. Student attendance shall be consistent with University policy.

Professionalism and Academic Integrity Policy

Students are expected to uphold <u>UNK Values</u> by being engaged in the course, conducting yourself with integrity, and treating others with respect and inclusivity. Critical questions and discussion to advance knowledge and understanding are strongly encouraged but must be done in a respectful way. Students who habitually disturb the class and have been warned may suffer a reduction in their final class grade.

Academic honesty is essential to the existence and integrity of an institution of higher education. The responsibility for maintaining that integrity is shared by all members of the academic community. To further serve this end, the University of Nebraska at Kearney has a policy relating to academic integrity. To ensure all students understand the expectations, UNK has adopted Academic Integrity Policy.

"Academic Integrity" is a term that encapsulates honesty, trust, fairness, respect, and responsibility among students and faculty. You are expected to follow the <u>University of Nebraska Student Code of Conduct</u>. In particular,

"Students are expected to approach and complete their academic work with integrity. They are expected to do their own work, to be honest in the statements they make, to refrain from harming others, to refrain from improperly helping others, and to follow the rules. Students must read instructions and syllabi carefully so that they know what their instructors expect in terms of academic integrity.

Students who are unsure whether or not particular conduct is appropriate should ask their instructors or university administrators. Failing to act with integrity is a violation of the Code." (Student Code of Conduct, Section IIA)

You may be asked to affirm the statement as true on submitted work: "By submitting this test or assignment, I unequivocally state that all work is entirely my own and does not violate UNK's Academic Integrity policy."

Plagiarism: It is of utmost importance in this course to understand and avoid plagiarism. Writing discussion posts and a paper critique are a core feature of this course. TurnItln may be used for assignments submitted in Canvas. For more information and tips, please visit the TurnItln's webpage "Preventing Plagiarism when Writing" or reach out to me for guidance. If you plagiarize in your submitted work you could fail the assignment or fail the course. Each instance of plagiarism, classroom cheating, and other types of academic dishonesty will be addressed in accordance with the UNK Academic Integrity policy.

University Policies and Resources

All students at the University of Nebraska Kearney should be aware of the following universitywide course policies and resources.

Attendance Policy

Your instructor may have indicated on their syllabus an attendance policy specific to their class. If so, that is the policy with which you must comply. If no other policy is stated, the University-wide attendance policy will apply. You can find the <u>Student Attendance Policy Statement online</u>.

Reporting Student Sexual Harassment, Sexual Violence or Sexual Assault

Reporting allegations of rape, domestic violence, dating violence, sexual assault, sexual harassment, and stalking enables the University to promptly provide support to the impacted student(s), and to take appropriate action to prevent a recurrence of such sexual misconduct and protect the campus community. Confidentiality will be respected to the greatest degree possible. Any student who believes she or he may be the victim of sexual misconduct is encouraged to report to one or more of the following resources:

Local Domestic Violence, Sexual Assault Advocacy Agency 308-237-2599

Campus Police (or Security) 308-865-8911

Title IX Coordinator 308-865-8655

Retaliation against the student making the report, whether by students or University employees, will not be tolerated.

Students with Disabilities

It is the policy of the University of Nebraska at Kearney to provide flexible and individualized reasonable accommodation to students with documented disabilities. To receive accommodation services for a disability, students must be registered with the UNK Disabilities Services for Students (DSS) office, 175 Memorial Student Affairs Building, 308-865-8214 or by email unkdso@unk.edu

Students Who are Pregnant

It is the policy of the University of Nebraska at Kearney to provide flexible and individualized reasonable accommodation to students who are pregnant. To receive accommodation services due to pregnancy, students must contact the Student Health office at 308.865.8218. The following links provide information for students and faculty regarding pregnancy rights. https://thepregnantscholar.org/title-ix-basics/

https://nwlc.org/resource/faq-pregnant-and-parenting-college-graduate-students-rights/

UNK Statement of Diversity & Inclusion

UNK stands in solidarity and unity with our students of color, our Latinx and international students, our LGBTQIA+ students and students from other marginalized groups in opposition to racism and prejudice in any form, wherever it may exist. It is the job of institutions of higher education, indeed their duty, to provide a haven for the safe and meaningful exchange of ideas and to support peaceful disagreement and discussion. In our classes, we strive to maintain a positive learning environment based upon open communication and mutual respect. UNK does not discriminate on the basis of race, color, national origin, age, religion, sex, gender, sexual orientation, disability or political affiliation. Respect for the diversity of our backgrounds and varied life experiences is essential to learning from our similarities as well as our differences. The following link provides resources and other information regarding D&I: https://www.unk.edu/about/equity-access-diversity.php

BIOL827: Biological Statistics

Tentative Fall 2022 Course Schedule

All work due by 11:59PM CT unless otherwise indicated. Required content not listed here may be provided in Canvas.

Module 1 Introduction to Statistics

Module 1 1	illioduction to Statistics	
22 Aug	Module begins Mini lectures (4) Textbook (1) Journal article (1)	Chapter 1
Due 23 Aug 25 Aug 27 Aug	Assignment Discussion Discussion Quiz	Leader posts article and questions Initial response
28 Aug 29 Aug 29 Aug	Discussion Problem set Module ends	Replies to groupmates

Module 2 Data Management

29 Aug	Module begins	
	Mini lectures (4)	
	Textbook (0) Journal articles (1) Websites (3) Optional materials (6)	No assigned textbook readings
	Optional materials (0)	

Due	Assignment	
30 Aug	Discussion	Leader posts article and questions
1 Sep	Discussion	Initial response
3 Sep	Quiz	·
5 Sep	Discussion	Replies to groupmates
5 Sep	Problem set	
5 Sep	Module ends	

Module 3 Describing and Visualizing Data

5 Sep	Module begins Mini lectures (4) Textbook (2) Journal article (1) Booklet (1)	Chapters 2 and 3
Due 6 Sep 8 Sep 10 Sep 11 Sep 12 Sep 12 Sep	Assignment Discussion Discussion Quiz Discussion Problem set Module ends	Leader posts article and questions Initial response Replies to groupmates

12 Sep Module begins
Lectures (2)
Textbook (2) Chapters 4 and 5
Optional video (1)
Web interactive (1)

Due Assignment Discussion 13 Sep Leader posts article and questions 15 Sep Discussion Initial response 17 Sep Quiz Discussion Replies to groupmates 18 Sep 19 Sep Problem set Module ends 19 Sep

Module 5 Hypothesis testing

19 Sep Module begins
Mini lectures (4)
Textbook (1)
Journal article (1)

Chapter 6

Assignment Due Discussion 20 Sep Leader posts article and questions 22 Sep Discussion Initial response Unit 1 Exam Timed 24 Sep 25 Sep Discussion Replies to groupmates Unit 1 Exam Problem set 26 Sep 26 Sep Module ends

Module 6 Experimental Design

26 Sep Module begins Mini lectures (3) Textbook Chapter 14 Due Assignment 27 Sep Discussion Leader posts article and questions 29 Sep Discussion Initial response 1 Oct Quiz 2 Oct Discussion Replies to groupmates 3 Oct Problem set 3 Oct Module ends

Module 7 Binomial Data and the Binomial Distribution

3 Oct	Module begins Mini lectures (4) Textbook (3)	Chapter 7
Due 4 Oct 6 Oct 8 Oct	Assignment Discussion Discussion Quiz	Leader posts article and questions Initial response
9 Oct	Discussion	Replies to groupmates

10 Oct Problem set

10 Oct Module ends

Module 8 Categorical Data and the χ^2 Distribution*

10 Oct Module begins Mini lectures (4)

Textbook (3) Chapters 8, 9

Due Assignment

11 Oct Discussion Leader posts article and questions

13 Oct Discussion Initial response

15 Oct Quiz

16 Oct Discussion Replies to groupmates

16 Oct Problem set*

17 Oct Module ends

Module 9 The Normal Distribution*

19 Oct Module begins

Mini lectures (4)

Textbook (2) Chapter 10, Chapter 13 sections 13.1-13.3

External video (1) Optional materials (3)

Due Assignment

19 Oct* Discussion Leader posts article and questions

20 Oct Discussion Initial response

22 Oct Quiz

23 Oct Discussion Replies to groupmates

24 Oct Problem set **24 Oct Module ends**

Module 10 One-sample and Paired t-Tests

24 Oct	Module begins
	Mini la atura (7)

Mini lectures (7)

Textbook (3) Chapter 11, Chapter 12 sections 12.1-12.2,

Chapter 13 sections 13.4 and 13.7

Due Assignment

25 Oct Discussion Leader posts article and questions

27 Oct Discussion Initial response

29 Oct Quiz

30 Oct Discussion Replies to groupmates

31 Oct Problem set **31 Oct Module ends**

Module 11 Two-sample t-Tests

31 Oct Module begins

Mini lectures (5)

Textbook Chapter 12, Chapter 13 sections 13.5-13.7

^{*}Due date shifted to Sunday, 10/16, due to Fall Break. No late penalty if submitted by 10/17.

^{*}Dates shifted for Fall Break. Discussion leader due to post article & questions on Wednesday.

Journal article (1) Optional interactive (1)

DueAssignment1 NovDiscussionLeader posts article and questions3 NovDiscussionInitial response

5 Nov Unit 2 Exam Timed 6 Nov Discussion

6 Nov Discussion Replies to groupmates

7 Nov Unit 2 Exam Problem set

7 Nov Module ends

Module 12 Introduction to ANOVA

7 Nov Module begins

Mini lectures (5)

Textbook Chapter 15, Interleaf 8

Optional materials (2)

Due Assignment

8 Nov Discussion Leader posts article and questions

10 Nov Discussion Initial response

12 Nov Quiz

13 Nov Discussion Replies to groupmates

14 Nov Problem set **14 Nov Module ends**

Module 13 Complex ANOVA

14 Nov Module begins

Mini lectures (5)

Textbook Chapter 18

Journal article (1) Same as "optional" article in Module 11

Due Assignment

15 Nov Discussion Leader posts article and questions

17 Nov Discussion Initial response

19 Nov Quiz

20 Nov Discussion Replies to groupmates

21 Nov Problem set
21 Nov Module ends

Module 14 Correlation*

21 Nov Module begins

Mini lectures (6)

Textbook Chapter 16

Due Assignment

28 Nov* Quiz

5 Dec* Problem set Correlation questions included in Mod 15 Problem Set

28 Nov Module ends

^{*}Due dates shifted for Thanksgiving Break; Modules 14 and 15 overlap. No Mod 14 discussion.

Module 15 Regression

28 Nov Module begins

Mini lectures (6)

Textbook Chapter 17

Due Assignment

29 Nov Discussion Leader posts article and questions

1 Dec Discussion Initial response

3 Dec Quiz

4 Dec Discussion Replies to groupmates

5 Dec Problem set5 Dec Module ends

Module 16 Meta-analysis

5 Dec Module begins

Mini lectures (3)

Textbook 2nd Ed. Chpt 21 (available on eReserves), Interleaf 10

Journal articles (3) Optional materials (1)

Due Assignment

6 Dec Discussion Leader posts article and questions

8 Dec Discussion Initial response

10 Dec Quiz

12 Dec

11 Dec Discussion Replies to groupmates

12 Dec Problem set
12 Dec Module ends

Module 17 Final Exam Week

Module begins

Due	Assignment	
13 Dec	Final Exam Critique	Due by 11:59PM CST
14 Dec	Final Exam Timed	Due by 11:59PM CST
15 Dec	Final Exam Problem Set	Due by 5:00PM CST
15 Dec	Course ends	-