

THE EFFECT OF ALMOND PHYTOCHEMICALS ON CANCER SUSCEPTIBILITY IN PLANARIA

LING XU

ORIGINAL QUESTION AND HYPOTHESIS



How do protocatechuic acid and catechin impact the growth of tumors in planaria exposed to carcinogens (benzo(a)pyrene and cadmium solution)?



It is hypothesized that planaria introduced to both protocatechuic acid and catechin before the exposure of carcinogens will develop little to no tumors compared to specimen solely exposed to carcinogens.

ADJUSTED QUESTION AND HYPOTHESIS



How do protocatechuic acid and catechin impact the survival rate of planaria exposed to carcinogens (benzo(a)pyrene and cadmium solution)?



It is hypothesized that planaria introduced to both protocatechuic acid and catechin before the exposure of carcinogens will result in the highest survival rate of planaria specimen.

BACKGROUND



Phagocata gracilis (black planaria) is a type of flatworm with regenerative properties



Benzo(a)pyrene and cadmium solution are known human carcinogens (Group 1) that are also harmful to aquatic life; however, the substances can already be found in the current environment such as grilled meats, tobacco smoke, tar, pesticides etc.



Protocatechuic acid and catechin are phytochemicals that are present in common foods like almonds and antioxidative products like green tea

Independent variables: type of chemical to which planaria is exposed

Categories include:

Control:
spring
water
environment

Exposure to
only the 2
carcinogens

Exposure to
only the 2
phytochemicals

Exposure to only
protocatechuic
acid

Exposure to
only catechin

Exposure to
protocatechuic
acid before
introducing the 2
carcinogens

Exposure to
catechin
before
introducing
the 2
carcinogens

Exposure to
both
phytochemicals
before
introducing the
2 carcinogens

Dependent variable: the survival rate of planaria

*although originally planned to measure tumor growth in planaria, the specimen were found to perish quickly before developing neoplastic nodules

VARIABLES

MATERIALS



Cadmium solution

- 1 COMPOUND MICROSCOPE
- 16 PETRI DISHES (25X100MM, 5ML)
- 100 PLANARIA SPECIMEN
- 75 ML PROTOCATECHUIC ACID
- 75 ML CATECHIN
- 4 SYRINGES (EACH WITH A CAPACITY OF 5ML)
- 100 UG/ML OF BENZOPYRENE SOLUTION
- 75 ML OF CADMIUM STANDARD SOLUTION
- 1 FACEMASK AND SAFETY GOGGLES
- DISPOSABLE PLASTIC GLOVES
- SPRING WATER
- 1 MICRO-SCOOP

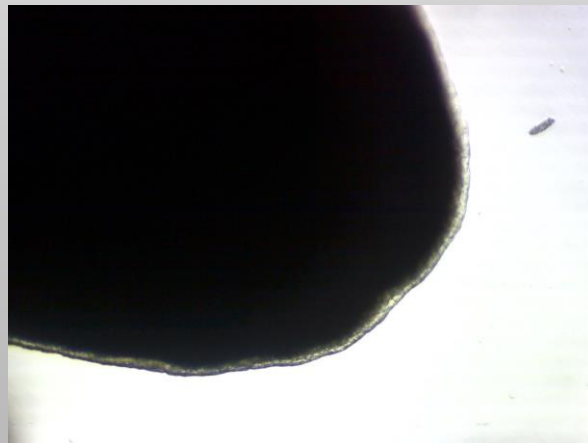
PROCEDURE



1. LABEL EACH PETRI DISHES (2 PETRI DISHES PER CATEGORY) WITH ONE OF 16 CATEGORIES IN WHICH PLANARIA ARE EXPOSED TO DIFFERENT SUBSTANCES, AND PLACE PETRI DISHES IN A LOCATION WITH MINIMAL SUNLIGHT IN THE SCHOOL LAB
2. SEPARATE 5 PLANARIA SPECIMEN INTO EACH PETRI DISH FILLED WITH SPRING WATER
3. ADD 0.05 ML OF BENZOPYRENE AND 0.05 ML CADMIUM SOLUTION ONLY INTO TWO PETRI DISHES OF PLANARIA EVERY DAY FOR 4 WEEKS.
4. EXPOSE TWO OTHER PETRI DISHES OF PLANARIA TO 0.1 ML OF PROTOCATECHUIC ACID ONLY EVERY DAY
5. REPEAT STEP 4 WITH 0.1 ML OF CATECHIN.
6. REPEAT STEP 4 WITH 0.05 ML OF PROTOCATECHUIC ACID AND 0.05 ML OF CATECHIN EACH DAY



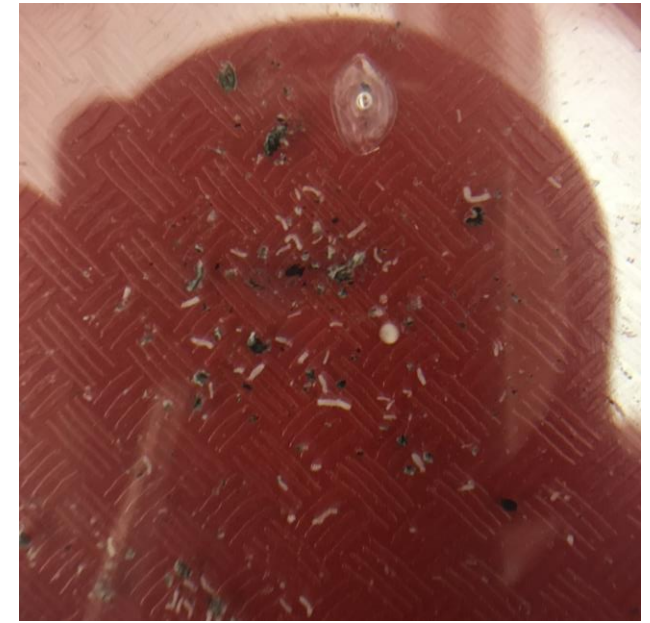
PROCEDURE (CONTINUED)



7. ADD 0.1 ML OF PROTOCATECHUIC ACID TO TWO ADDITIONAL PETRI DISHES FOR EACH DAY FOR 72 HOURS.
 - α. AFTER 72 HOURS, THE PLANARIA WILL BE EXPOSED TO 0.05 ML OF BENZOPYRENE AND 0.05 ML OF CADMIUM SOLUTION FOR 4 WEEKS.
8. ADD 0.1 ML OF CATECHIN TO TWO OTHER PETRI DISHES EACH DAY FOR 72 HOURS
 - α. REPEAT STEP 7A
9. ADD 0.05 ML OF PROTOCATECHUIC ACID AND 0.05 ML OF CATECHIN TO TWO PETRI DISHES EACH DAY FOR 72 HOURS
 - α. REPEAT STEP 7A
10. OBSERVE PLANARIA BEHAVIOR EVERY OTHER DAY FOR THE ENTIRE COURSE OF THE EXPERIMENT WITH THE MICROSCOPE
 - α. RECORD THE NUMBER OF PLANARIA TO PERISH
11. REMOVE ALL MATERIALS FROM SCHOOL AND DISPOSE APPROPRIATELY UPON TERMINATION OF THE EXPERIMENT

PLANARIA BEHAVIOR

- WHILE ORIGINALLY HOPING TO OBSERVE TUMOR GROWTH OF PLANARIA, THE PLANARIA WERE FOUND TO PERISH IN A QUICK PROGRESSION WITHOUT THE DEVELOPMENT OF NEOPLASTIC NODULES



- THE PROCEDURE WAS ADJUSTED TO ACCOMMODATE THIS OBSERVATION TO MEASURE SURVIVAL RATE RATHER THAN TUMOR DEVELOPMENT

DATA RESULTS

- THE EXPERIMENT WAS RECONDUCTED AFTER DAY 5 WHERE ALL PLANARIA EXPOSED TO THE PHYTOCHEMICALS HAD DISINTEGRATED
 - IT WAS INFERRED THAT THE PLANARIA COULD NOT TOLERATE THE CONCENTRATION OF PHYTOCHEMICALS
 - THE PROCEDURE WAS ADJUSTED TO LOWER THE CONCENTRATION OF CHEMICALS TO 0.1 ML INSTEAD OF 0.25 ML PER DAY



Planaria exposed
to catechin on
Day 5

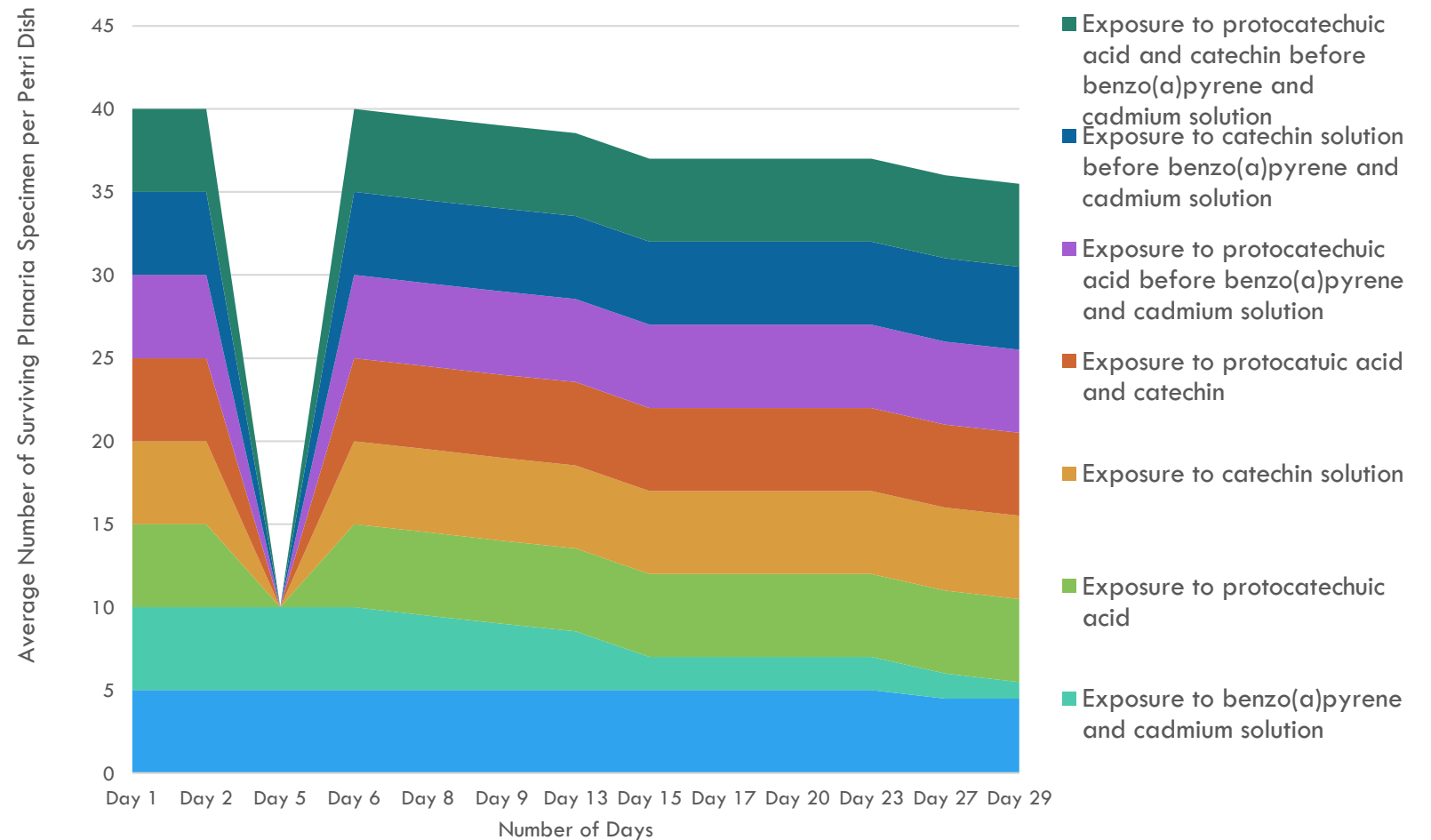
Data Chart #1: The Number of Surviving Planaria Specimen per Petri Dish When Exposed to Various Chemicals

	Control (spring water environment)		Exposure to benzo(a)pyrene and cadmium solution		Exposure to protocatechuic acid		Exposure to catechin solution		Exposure to protocatechuic acid and catechin solution		Exposure to protocatechuic acid before benzo(a)pyrene and cadmium solution		Exposure to catechin solution before benzo(a)pyrene and cadmium solution		Exposure to protocatechuic acid and catechin solution before benzo(a)pyrene and cadmium solution	
Day 1	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
Day 2	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
Day 5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Day 6	5	5	4	5	5	5	5	5	5	5	5	5	5	5	5	5
Day 8	5	5	4	5	5	5	5	5	5	5	5	5	5	5	5	5
Day 9	5	5	4	4	5	5	5	5	5	5	5	5	5	5	5	5
Day 13	5	5	3	4	5	5	5	5	5	5	5	5	5	5	5	5
Day 15	5	5	3	4	5	5	5	5	5	5	5	5	5	5	5	5
Day 17	5	5	0	4	5	5	5	5	5	5	5	5	5	5	5	5
Day 20	5	5	0	4	5	5	5	5	5	5	5	5	5	5	5	5
Day 23	5	5	0	4	5	5	5	5	5	5	5	5	5	5	5	5
Day 27	5	4	0	3	5	5	5	5	5	5	5	5	5	5	5	5
Day 29	5	4	0	2	5	5	5	5	5	5	5	5	5	5	5	5

DATA RESULTS

DATA ANALYSIS

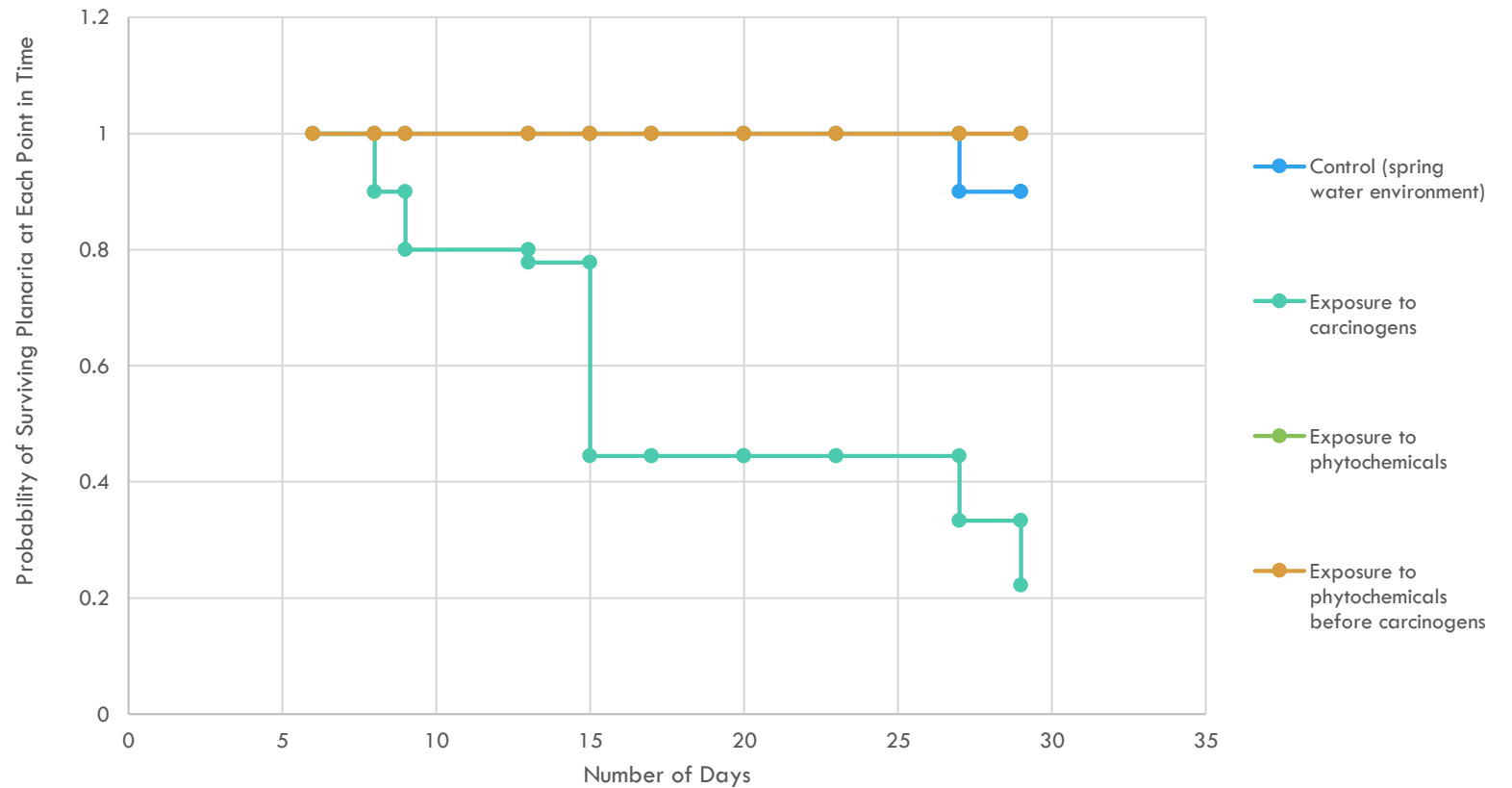
Graph #2: The Number of Surviving Planaria When Exposed to Various Chemicals



SURVIVAL ANALYSIS

- BY LUMPING SUBGROUPS, SAMPLE SIZE CAN BE INCREASED TO LIMIT VARIABILITY

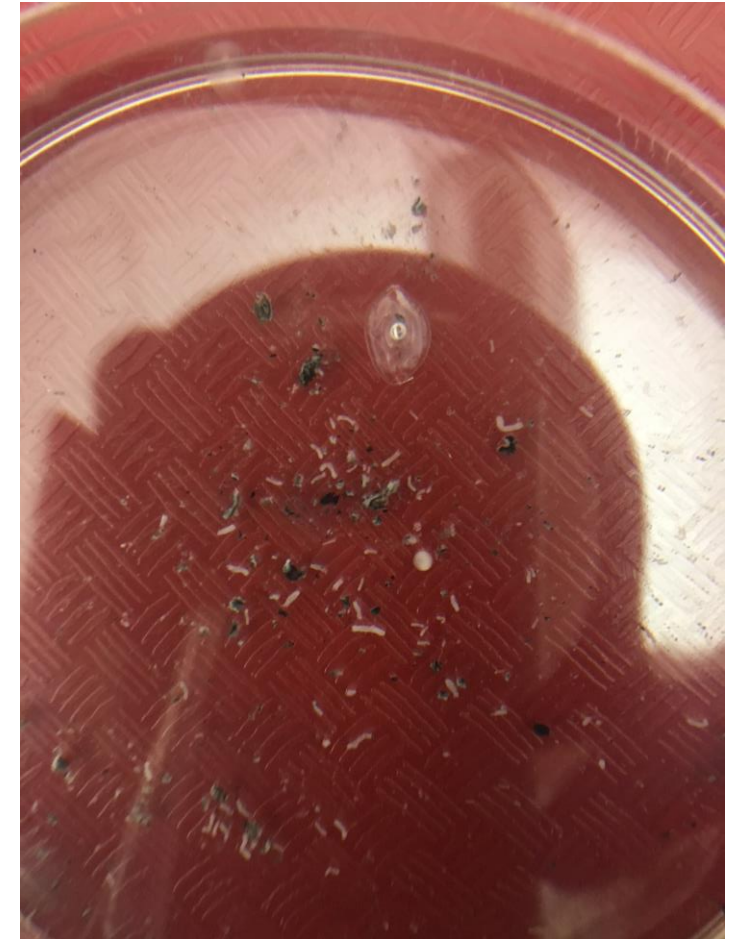
Kaplan-Meier Survival Rates of Planaria Exposed to Various Chemicals Beginning at Day 6



COMPARISON



Planaria exposed to
protocatchuic acid before
carcinogens



Planaria exposed to only
carcinogens

CONCLUSION AND POTENTIAL IMPACT

- **HYPOTHESIS WAS CONFIRMED:**
 - WHILE PLANARIA ONLY EXPOSED TO BENZO(A)PYRENE AND CADMIUM SOLUTION HAVE DIED OVER TIME (DECREASING SURVIVAL RATE), THOSE WHO WERE EXPOSED TO PROTOCATECHUIC ACID AND/OR CATECHIN BEFORE THE INTRODUCTION OF CARCINOGENS HAVE NOT PERISHED AT ALL
 - EVEN WHILE PLANARIA FROM THE CONTROL GROUP DIED, PLANARIA EXPOSED TO PHYTOCHEMICALS DID NOT
 - PROTOCATECHUIC ACID AND CATECHIN IN SMALL QUANTITIES (APPROX. 0.1 ML) POSE BENEFITS TO THE PLANARIA'S BODILY SYSTEMS
- **THE RESULTS CAN BE APPLIED TO HUMANS AS WELL**
 - WHILE PHYTOCHEMICALS CANNOT BE NECESSARILY UTILIZED AS A TREATMENT METHOD, CONSISTENT CONSUMPTION OF FOODS HIGH IN ANTIOXIDANTS MAY LOWER RISKS OF CANCER DEVELOPMENT IN THE LONG TERM

RELEVANCE TO SOCIETY



Common treatments for cancer include chemotherapy and other high-risk drugs that often pose serious side effects and harm vital bodily functions in the process of attempting to destroy the cancerous cells



Instead of attempting to eradicate the tumors, phytochemicals may reduce the risk of mutations in the cell cycle that results in cancer

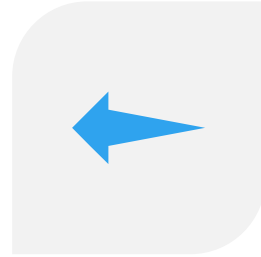


Phytochemicals (specifically procatechuic acid and catechin) that can be found in every-day markets such as almonds may possess the ability to lower the risk of the proliferation of cancer

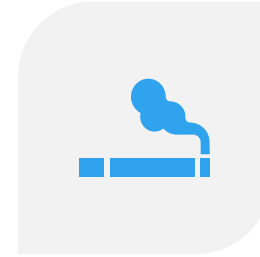


A window into a possible research field

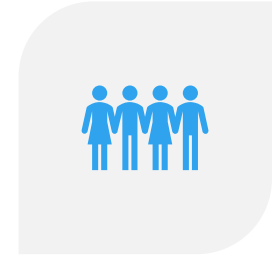
ERRORS AND FUTURE REVISIONS



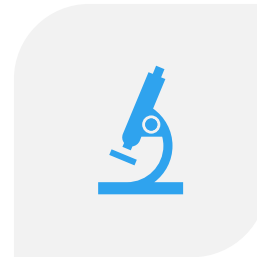
SMALLER
CONCENTRATIONS OF
PHYTOCHEMICALS



UTILIZE Milder
CARCINOGENS



EMPLOY A LARGER
SAMPLE SIZE



UTILIZE DIFFERENT
SPECIES OF
PLANARIA



TEST EXPOSURE TO
PHYTOCHEMICALS
AFTER CARCINOGEN
INTRODUCTION