Original Research Article

Field Code Changed

Hypothetical theoretical model of experiment for Sequential repeat environment adaptation of Commensal bacterial, transplantation a changing neurological behavior of *Drosophila*

Abstract:

A hypothesis theory is presented by using game theory in this research paper. According to which 3 different species (species A and species B) are passed through the system of different compartments. Both species can only interact in compartment E and the bacterial species experiences a particular environment as it passes through each compartment. Since only compartment E is the location of both battery species interaction, and is the location of cooperation for both Bacterial species.

If compartment E contains a limited amount of essential carbon source like sucrose for the bacteriological system, it is likely that species A and species B initially compete, but frequent contact with each other in sequence turned corporations into compartment E for limited self-uptake of sucrose. Competition was reduced so that it is also available for other species.

This adaptive behaviour of Bacterial system can be confirmed through CAFE Assay by transferring Adapted bacteria to *Drosophila.*

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Comment [HT1]: Rewrite abstract

Comment [HT2]: Check it, 2 or 3 species

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If we practice any process and repeating that same process multiple times, we become mastered and get adapted to do that process very easily by applying strategy learned in doing them so that fewer efforts were needed. Similarly, if bacterial cell pass through the repeated sequential environment condition and allow to interact with other bacteria in specific environmental niche, only after passing through sequential environmental condition that bacterial cell start adapting or set a rhythm for specific gene expression to survive and interact with other bacterial cell and become a co-operator/defector for other bacterial cells in specific environment only. So, it is only possible to become a mutant compared to wild-type organism passing through genetic mutation like SNPs, or phenotypic heterogeneity compared to the wild-type not going through this pevolvic conditional adaptation. So, it can be possible that mutant has a deferent heat map of whole gene expression profile compared to wild-type and it is easy to find out that gene particularly involved in cooperation behavior toward the other bacterial cell. If this hypothesis was correct then it will be beneficial to study conditional adaptive cooperation behavior among bacterial cells. After rhythmic setup in bacterial co-operation toward the other organisms, it become quite useful and interesting to know that how organism behave in its specific environment while interacting with other organisms, it has been observed that removal of co-operator/defector from interacting zone area after multiple encountering of the same process, change in genetic, phenotypic and behavioral response is expected to obtain from co-operator or defector in absence of partner group behavioral organism.

So the questions arise ~~W~~what happens if we transplant conditional adapted co-operator mutant derived beneficial mutation like- SNPs or modify the gene expression of a specific gene in wild-type organism? Can we magnify the cooperation group behavior in bacteria by either applying clonal interference/Moran model?

***Materials and Methods*:**

prisoner's dilemma (IPD) scoring matrix for calculation of mean fitness of organism after and before cooperation. and By Game theory method use to find development of altruism behavior .

***Results*:** The same type of recurring environment~~ will~~ set the rhythm in the bacterial system, and one particular particle environment ~~will~~ <u>was</u> prepare<u>d</u> specials as adaptive acceptors for co-operative adaptation in the bacterial system.

Conclusion:

This theoretical model will help to understand the effect of repeated sequential environments on cooperative behavioural adaptation. Which will help us in the future to implant an adaptive trait in the bacterial system.

***Key Word*:**
~~c~~<u>C</u>*omputer*-based iterated prisoner's dilemma (IPD) scoring matrix; Moran model, capillary feeder (CAFE) assay, Prisoner's Dilemma, Clonal interference model, Game theory method

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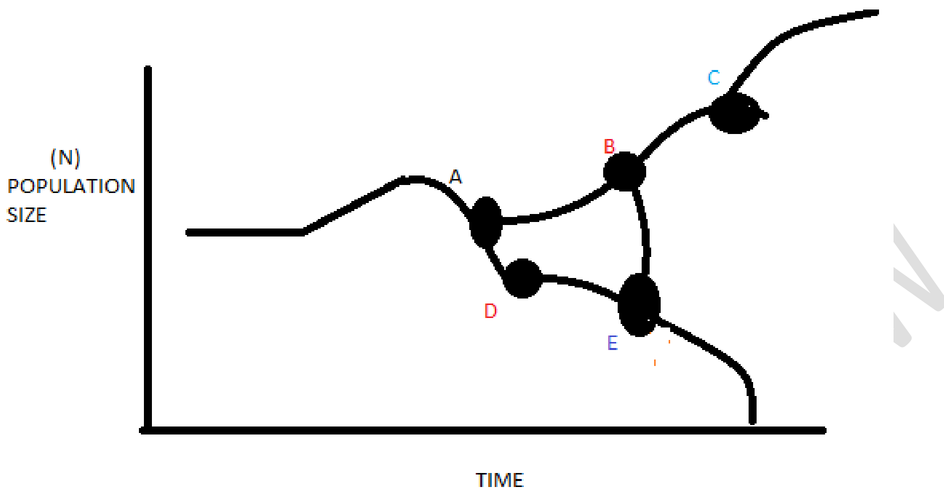
I. Introduction

Different environmental conditions like- pH, temperature, specific light source, magnetic field, etc; affects a lot in cell physiological behavior, developmental process, sex determination, migration behavior & differentiation process, etc. These all processes are sensed by various signaling molecules leading to signaling cascade interconnected with each other and even some were experimentally proved which says that cell sense the environment and apply the strategy to sense the specific component but when environment changes it is changing the sensing strategy due to disappearance of that desired component from the environment which is responsible for the signaling cascade. Such conditions, within bacterial cell setup their biological signaling cascade and gene expression circuits called pevolvic condition. It is similarly applied during training of pet animals for example - in case of a dog, we train them by giving food calling by bell ringing sign, etc.

It is well known that when an individual bacterial cell or an organism comes in contact with other bacterial cells they start interacting with other individuals in the population. The interaction can be positive or negative or an individual may act as a co-operator or defector depending upon the situation. For example, by sensing internal iron deficiency, *Pseudomonas aerogenosa* who have the ability to produce siderophore start producing it and excrete out in the environment, it acts as a quencher for iron and again engulfed by bacterial cell through the phenomenon of endocytosis<u>,</u> which become beneficial for other bacterial cells in the population as it decreases the cost of bacterial cell for producing siderophore. Thus, it was positive kind of behavior among bacteria population. But when an amount of iron decreases in the external environment, the ability to produce siderophore by both groups of population lead to competition. In that case, population migrate from positive group behaviour to compositional behavior so that both cells learn siderophore producing strategy which is not an evolutionary stable strategy (ESS). Suppose if one cell of the population start producing siderophore (mention by A+) and another one is siderophore deficient (mention by B-), such conditions maintain a stable state of siderophore level in the environment.

In different phases of cell growth and development, cell exposes different environmental conditions where some may be beneficial or some are harmful, in such conditions, various mutation occur in their genome which starts spreading into the population during cell maturity when it starts producing progeny cells. An accumulated mutation can be beneficial or harmful. If it's harmful, it decreases the mean fitness of the population which can be identified by Moran mathematical model useful in clonal cell probability of beneficial mutant to spread into the population by dividing of a cell or by any other medium.

In Clonal interference model, each bacterial cell act as an individual clone so if any mutation arises in the clone, it also spreads in the upcoming daughter cell. If a mutation is beneficial, it increases the mean fitness of a group of the population. Arising sub-sequential mutation is a beneficial mutation in the same clonal cell leading to more enhancement in the fitness of a group of organism in the population because both separate mutation act as a separate mode of action for enhancing fitness by interference means result, which ultimately leads to increase in the number of group of organism and fitness in the population. Similarly, opposite accumulation of harmful mutation decreases the fitness of the group.



Graph 1 : Model graph

We ~~will~~ calculate the probability, for each beneficial mutation to fix into the population by using Moran model.

$$X= 1-1/r/1-1/r^{N}$$

Where N=Population size

and, an x= probability of fixation of the single mutation in a population with real fitness r

In this model, (x) fixation probability of benefit mutant either become a neutral mutation 1/N or it becomes a beneficial fixation mutant, defines as S. and, should be S>0, SN>>1.

By applying computational approach and a mathematical model equation of mutation rate (μ) in the population. Theoretically, we can calculate the time required to establish the mutation in the population by using following equation. This equation gives an idea of the time required to establish the mutation into the individual clone. It's also denoted by Test.

(Test.) Tmut.= 1/μns

As a cell grows and initiate division, it establishes beneficial mutation which starts spreading in the population, and time required to spread this mutation into the population is known as 'time for mutation fixation' or Tfix.

Theoretically, if the time required to fix mutation in individual clone (Test) -is longer than the time required for a beneficial mutation to fix into the population (1/μns>>1/s ln (NS)), in such case, clonal interference is not observed. In short, in such condition one beneficial mutation in an individual clonal cell does not give additive effect on the population of the same organism, each beneficial mutation limited only up to individuals, they will not spread into the population.

II. Material And Methods

Two main methods are used to study the conditional based cooperation among the bacterial population-

1. By heat map of whole genome by specific expression of genes using computer-based iterated prisoner's dilemma (IPD) scoring matrix for calculation of mean fitness of organism after and before cooperation.
2. By Game theory method, which is useful in interaction-based kind of studies. As we know that according to game theory rules, score distribution considering strategies of both co-operator and defector individuals relative to the population of A+ and B-. Generally, payout value finds higher in case of interaction between defector and co-operator individuals. A computer-based model of iterated prisoner's dilemma (IPD) is used to study the evolution of conditional base cooperation.

Suppose, Table 1 is an initial condition or first round of interaction, where cooperation is not evolved, in this case, we find following similar graph data, Fig.1.

Table.1 Initial iterated prisoner's dilemma (IPD)

	A+ POPULATION as co-operator (C)	B - POPULATION as defector (D)
A+ as co-operator	3	0
B- as defector	5	1

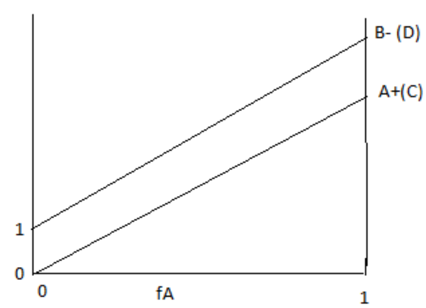


Fig.1 fractional cooperation by species A+

Results and discussion

Here in Fig.1, fA scoring matrices (0 to 1) indicate fractional co-operation by species A toward the species B.

But as interaction and co-operation get enhanced in specific environment, we may find change in score matrix, it gets increased from 0 to 1(Fig., 2, Ttable 2), the graph shows that as co-operation increase both line of payoff cross to each other, such situation allows calculating mean fitness of population and fractional cooperation by species A+.

Table. 2 Initial iterated prisoner's dilemma (IPD)

	A+ POPULATION as co-operator (C)	B- POPULATION as defector (D)
A+ as co-operator	5	0
B – as Defector	3	1

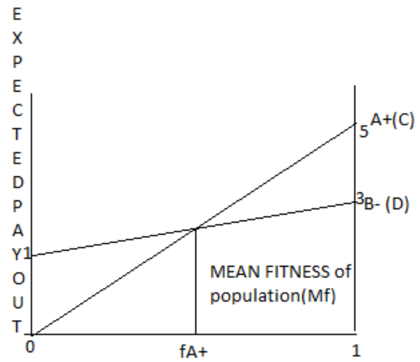


Fig.: 2 fractional cooperation by species A+

Procedure methodology

Experiment 1

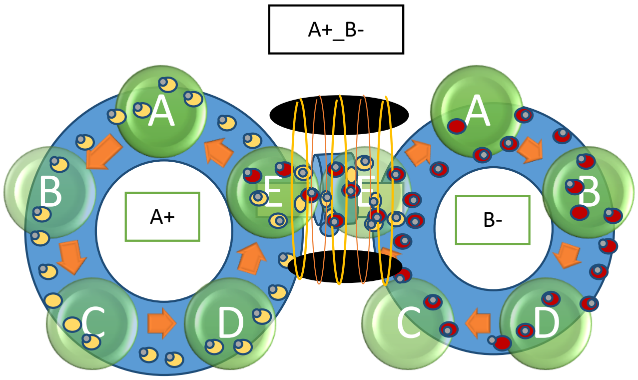


Fig.3 hypothetical experimental setup

Theoretically, hypothetical experimental setup is designed (Fig.3) to give a conditional environment to an organism to adapt to an artificial environment. According to Ffig.1 setup, A to E are individual compartments where each compartment has a different environmental condition like- different temperature, osmotic stress, light source and magnetic field, etc. In such artificial condition, the organisms are allowed to migrate only in one direction i.e- from A to E, where compartment E has more specific environment compared to other compartments like- the additional specificity of light source and the generation of magnetic field by interaction of the external magnet and magnetic particle that are transplanted into the organism to sense E compartment.

Hypothetically, It was believed that repeated multiple rounds of rotational interaction will start fixing rhythm gene expression pattern in the organism. This pattern starts fixing like 1,4,6 gene expression up-regulates in A compartment while 2,3,4 gene expression up-regulates in B compartment. Similarly, group cooperation related gene expression pattern set in E compartment. Like siderophore gene expression base mutation is only observed in E compartment.

This change occurred due to a mutation which can be structural, single nucleotide polymorphic (SNP's), or phenotypic in nature. The first step of this hypothetical experiment is collected for such mutation from E compartment organism gene expression pattern. Here, a collection of mutation done by comparing the whole genome of a control system with E compartment organism. Control system defines a random interaction between co-operator (A+) and Defector (B-) where A+/B- is not filling sequential conditional environment (fig.4).

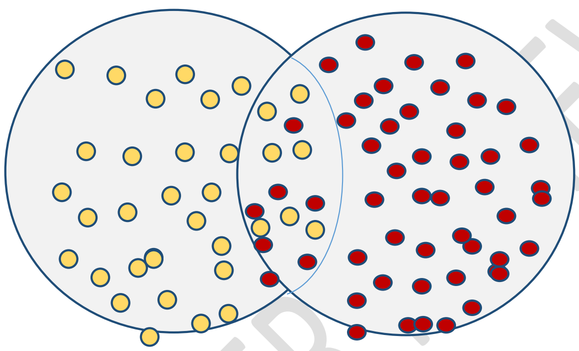


Fig.4. Control system

After finding such mutant gene, and checking it's probability for an establishment in the single clone fixation of such mutation into the group of a population by using Moran model. Once, confirmation is done by using computational and mathematical models using bioinformatic approach, that such mutant has a high probability to fix into the group of the population that will transplant all its mutant gene into the single clonal population and try to apply through clonal interference into the population. At the end of this experiment, the presence of all beneficial cooperation mutation in the group of the organism were checked in order for the success of enhanced co-operative interaction established among organisms or not?

Experiment 2:

If we decrease or increase specific condition of the environment in compartment E. Will it affect to set the rhythm toward the cooperation behaviour among the bacteria or not? According to current hypothesis, if we remove the magnetic field and light source from compartment E or in other words if we decrease specific environment for cooperation in compartment E, an organism requires more time to set the rhythm for cooperation and adapt in the environment, because decreasing specific environment in compartment E .Fig.5,6,7.

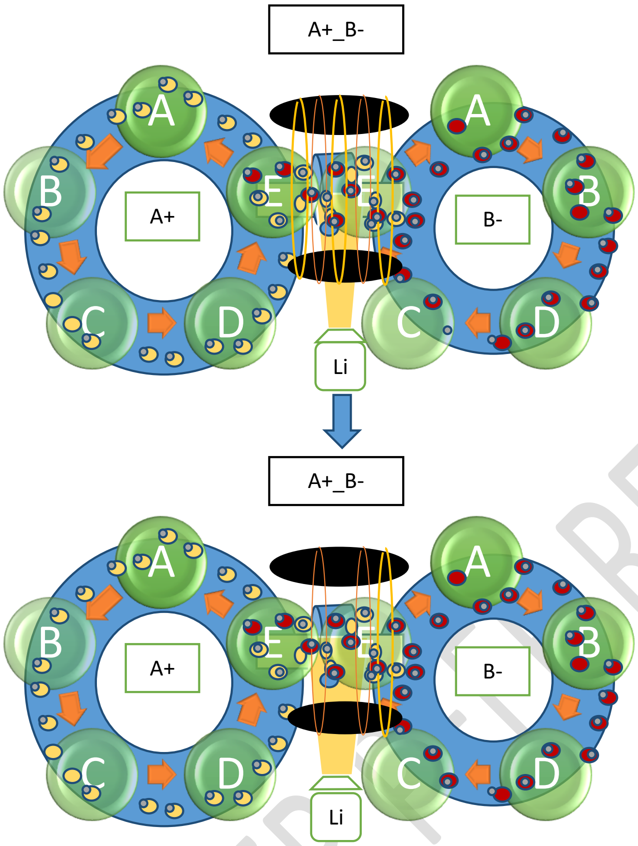


Fig.5 Removal of specificity of E compartment by, decreasing magnetic field.

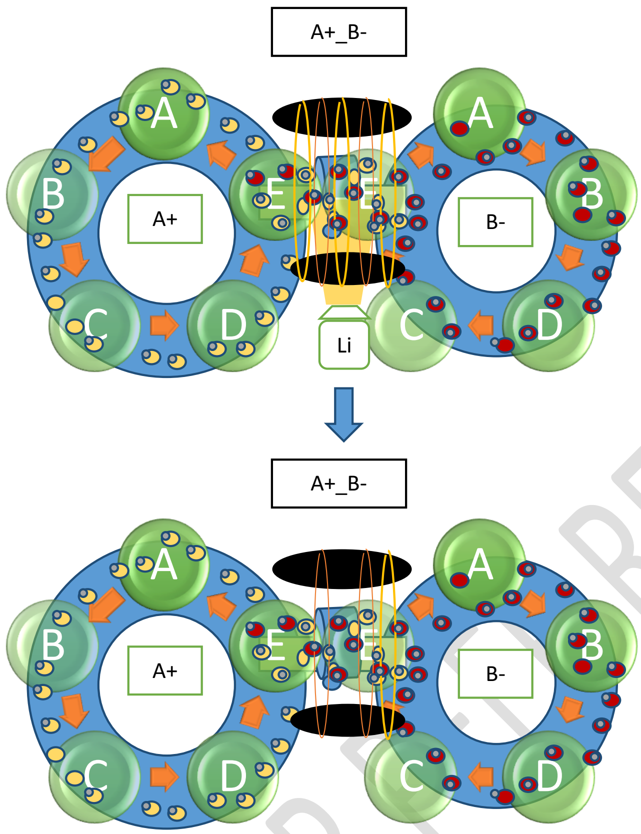


Fig.6 Removal of specificity of E compartment by decrease magnetic field and removal of the light source.

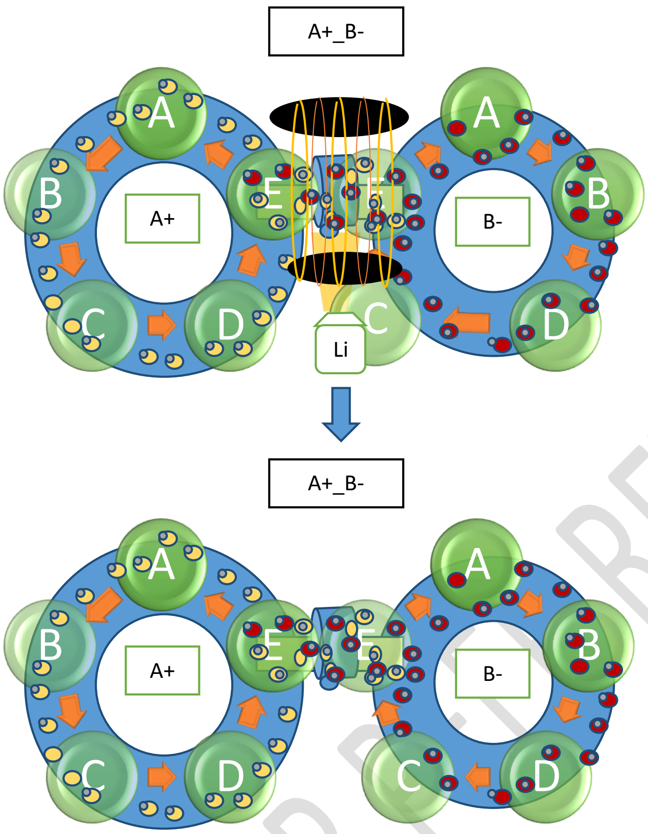


Fig 7. Removal of specificity of E compartment by escaping magnetic field and light source.

Experiment 3:

Once rhythm of gene expression set in A to E compartment after multiple rounds of interaction and suddenly if we remove the interaction between A+ and B-, Does it showed any drastic change in the behaviour of gene expression? or any essential/drastic mutation occurred for again setting rhythmic conditions among the bacterial population? (Fig.8).

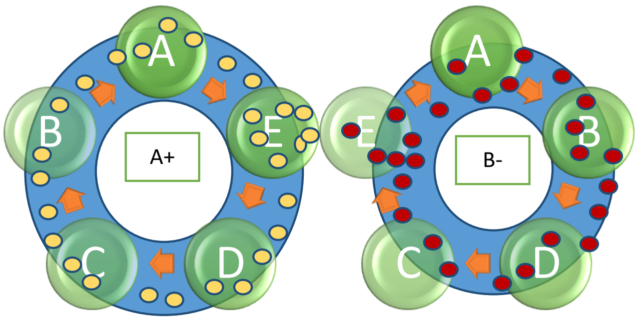


Fig.8. Removal of conditional interaction between A+ and B-, after multiple time interaction.

Experiment 4

Ricardo Leitao Goncealves *et al* research group hypothesis demonstrated that in absence of essential amino acid (eAA), *Drosophila melanogaster* attract towards the yeast extract (which is a rich source of protein and amino acid). But in presence of specific commensal bacteria as gut microbiota of drosophila, it changes feeding behaviour in capillary feeder (CAFE) assay, by unknown mechanism where commensal bacteria increases the feeding behaviour of drosophila towards sucrose capillary action and neutral response towards the yeast extract even if this holistic media lacking essential amino acid.

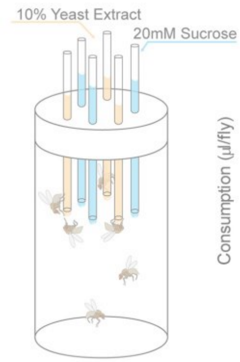


Figure 9 : (Ricardo Leitão-GoncËalves *et al.,* 2016)

STRATEGY

If conditional environment for adaptation to this commensal bacteria is provided to adapt in co-operation with other group of commensal bacteria in compartment E only, for sucrose uptake. In short, if condition is created in commensal organism for adaptation to co-operate with other commensal bacteria for limited available sucrose in E compartment , than after multiple round of interaction it leads to set a rhythm in bacterial cell to co-operate only in E compartment for sucrose uptake. The adaptation may decrease the uptake rate of sucrose by one group of commensal bacteria so that the other group can easily uptake sucrose in commensalism manner. After conditional co-operation rhythm set in the commensal bacteria, we are going to transplant such bacteria into drosophila and observe for the capillary feeding behavior for sucrose in each separate compartment from A to E using capillary feeder (CAFE) assay.

If this hypothesis proved correct, and rhythm was set for co-operation among commensal bacteria, they must modify drosophila behavior in such a way so that drosophila feed less amount of sucrose in compartment E compared to other compartments by CAFE assay because bacteria always co-operate in compartment E during the adaptation (fig.9).

For example- during adaptation, all compartments have very less amount of sucrose available and for survival of bacteria only sucrose is present in all compartments from A to E. So in compartment A to D, only A+ commensal bacteria are present but in compartment E when group of organism come in contact in such environment, they start co-operating with each other while trying to distribute sucrose within the population. If somehow after the transplant of such commensal bacteria into *Drosophila*, the sensing mechanism of E compartment environment remain same due to which drosophila must take less sucrose feed in compartment E compare to other compartment

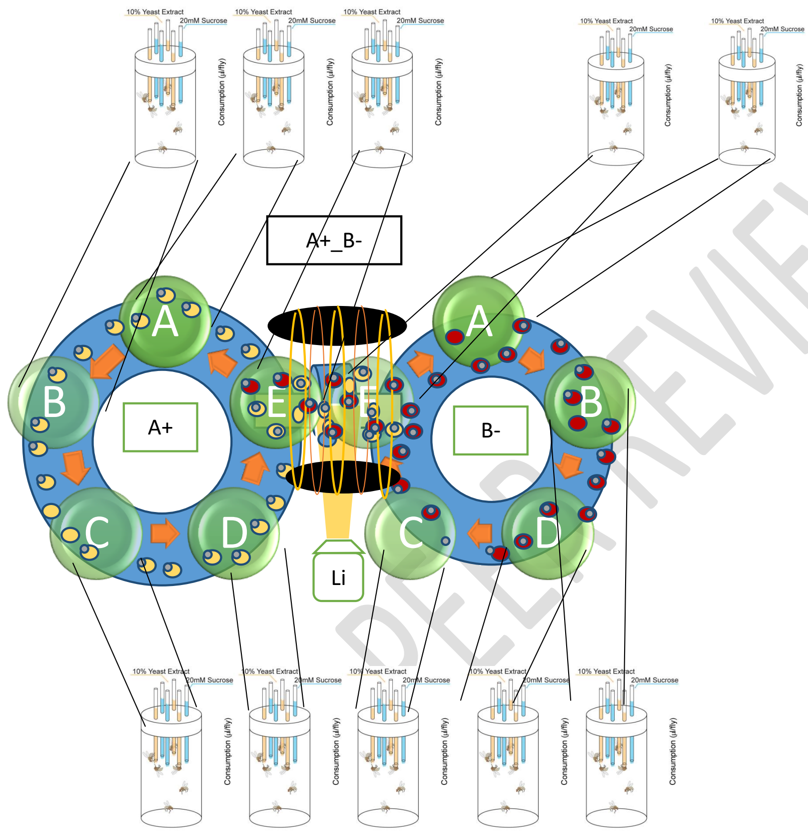


Fig. 10 Study protocol

III. Conclusion

Experiment: 1

Multiple rounds of interaction in specific sequence environment, lead to adaptation between co-operator/defector interaction. Thus, the conclusion can be made regarding that whether in such interaction organism has utilized the specific environmental condition for sensing and setting rhythm amongst each other or not. After finding multiple mutants in the repeated round of interaction between cooperator/defector compare to control and implant all mutant into the organism cooperator/ defector, whether it has magnified the cooperation behavior among the interacting bacterial population or not.

Experiment 2

Compartment E is more specific comparisons to other compartments due to which it has an additional environment like- the presence of magnetic field and specific light source. According to the hypothesis, an organism use this E compartment environment and senses and place his need to cooperate with another group of organism. The co-operator can utilize it as recognition mark for cooperation environment based on multiple time exposure given. If sequential removal of specific environment like- magnetic field and light source from conditional compartment E was done, it is going to affect the co-operation behaviour among a bacterial population.

Experiment 3

After multiple rounds of interaction and setting rhythm of gene expression among both group of organism. If suddenly, we stop interaction between co-operator/defector by separating both co-operator/defector system, whether still, they express the same gene responsible for co-operation with the same intensity or not can be concluded from this kind of behavior.

Experiment 4

After multiple rounds of interaction and setting rhythm of gene expression among both group of organism. If organism encounters same environments again, it starts to behave directly or indirectly same way or not? By recognizing the environmental condition.

References

Comment [HT4]: References are not cited in text, all should be cited accordingly

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