MS in General Biology Sample Comprehensive exam – Highly developed in all aspects

**Part 1: Journal Article Analysis**

Article #1

**Fruit Fall in Tropical and Temperate Forests: Implications for Frugivore Diversity.**

Goro Hanya, Shin-ichiro Aiba

**Problem: Is the diversity of frugivorous birds and primates affected by the amount of fruit fall in different regions?**

| Claim/Concept | Evidence/Support | Justification |
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| The difference in the amount of fruit fall between temperate and tropical regions could partially explain the difference in frugivore diversity between these regions. | * Fruit fall decreased from tropical to temperate forests with the exception of Australia. This indicates that fruit fall decreases with increasing latitude.<br>- Fruit fall in tropical forests was 454 ± 258 kg/ha/year, in temperate forests (excluding Australia) was 265 ± 227 kg/ha/year, and in temperate forests was 362 ± 352 kg/ha/year (Table 2a, Fig. 1)<br><br>* Fruit fall seemed to explain some of the variations in diversity of primates (Fig 3)<br>-The increased fruit fall in the tropics correlated with higher primate diversity in tropical regions (Fig 3) | There is more diversity of frugivores in tropical regions and this correlates with the increased amount of fruit fall that is seen in those tropical regions. Since the frugivores are using the fruit as a food source, it makes sense that if a larger amount of food is available that particular ecosystem can sustain a variety of organisms. Less fruit available in the temperate regions correlates with a lower diversity of frugivores in that region. Since there is a correlation between the amount of fruit fall and the diversity of the frugivores it is possible that this explains the difference in diversity between the temperate and tropical regions if only in part. |
| Fruit fall explains some of the variations in frugivore diversity between temperate and tropical regions but it is clear that other factors also | *The effect of fruit fall on diversity was different between primates and birds (Fig 3, Fig 4)<br><br>* No relationship between bird diversity and fruit fall was detected. (Fig 4) | Fruit fall in tropical forests was only 1.71 times larger than the fruit fall in temperate forests. This is smaller than the difference in frugivore diversity (pg 1088). Since we see a greater difference in diversity of frugivores than we see in the difference between the fruit fall between the two regions this would lead us to believe that fruit |

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| contribute to tropical regions containing higher frugivore diversity. | *Only temperate/tropical classification affected bird diversity (Fig 4). | fall contributes to the diversity difference between the two regions but it is not the only contributing factor. The correlation between fruit fall and frugivore diversity is probably found when looking at primates but not birds because primates tend to stay in a home range while birds have the opportunity to migrate to other food sources if needed. Ecosystems are complex and involve many factors weaved together. The diversity of frugivores seems to depend on fruit fall but also probably depends of factors such as the seasonality of fruit, the evolutionary history of the regions, and the availability of other food sources. |

Methodologies

The authors of this paper compared fruit fall between 53 sites ranging from the equator to the cool-temperate zone (36°S - 62°N) in Asia, Africa, North and South America, and Australia. (Table 1). A total of 25 tropical sites and 28 temperate sites were analyzed for this study. In order to get the data on these sites the authors combed through different literature and websites, they did not go to the sites themselves. Fruit fall was compared based on dry-weight, however when dry weight had not been recorded the authors estimated it at 29.5 % of the wet weights. Also if the data they were looking at had weighed the entire reproductive organs and not simply the fruit, the fruit weight was estimated at 63% of this.

The data was examined for 5 cases

- Entire
- Temperate and tropical excluding Australia
- Tropical
- Temperate
- Temperate excluding Australia

The relationships between latitude and fruit fall were examined using GSL regression.

Primate and bird diversity was obtained by reviewing the literature and include animals that are strictly frugivores, partial frugivores or granivores.

The GSL models for both birds and primates included

- Fruit fall
- Temperate/tropical classification
- Both fruit fall and temperate/tropical classification
- Both fruit fall and temperate/tropical classification as independent variables

Community College Connection

This paper would fit well in any ecology section of a biology class. It is a great example of the topic of interdependency of living things. All living things are affected and rely on the other living and nonliving components of their environment. Nothing exists in isolation. Factors that affect an organism in one way (ex: increase fruit fall leads to increased primate diversity) may not have the same effect on another organism (ex: no correlation between fruit fall and bird diversity). In an ecosystem, many factors tend to be in play at any single moment causing trickle down effects that are felt by many different organisms.

I would use this paper to help illustrate the point that sometimes factors in an environment can have a direct influence on an organism (amount of fruit fall on primate diversity) but other times these same factors do not directly influence other organisms (bird diversity). To teach this in the classroom I would split the students into groups and give them Figure 3 which shows a graph of the fruit fall vs the number of primate species. I would allow them to come to a conclusion on the effect of fruit fall on primate diversity. We would share out as a class and come to a consensus that the greater the fruit fall, the greater the primate diversity. I then would repeat this process with Fig 4 which shows the amount of fruit fall vs the number of bird species. Students should be able to come to a consensus that there is no correlation between increased fruit fall and increased bird diversity. Students would then be asked to come up with an explanation with their group on why they think this difference exists between primates and birds. They should also come up with a plan on what to study next to gain more insight into the differences. Groups would share their thoughts with the class. As a class we can talk about how many factors exist within an ecosystem and often times one factor might have greater influence over a particular organism but not another. In order to gain a complete understanding of an ecosystem one must study as many of these factors as possible.

Article #2

**Island hopping introduces Polynesian field crickets to novel environments, genetic bottlenecks, and rapid evolution.**

**Tinghitella et. al.**

**<u>Problem:</u> To identify the neutral processes that might influence sexual signal evolution in *Telegryllus oceanicus.***

| Claim/Concept | Evidence/Support | Justification |
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| *Telegryllus oceanicus* spread across the islands from Australia to Hawaii likely through the movements of Polynesian settlers (either intentionally or on their ships). | *Allelic richness decreases as you move west to east (From Australia to Hawaii). (Table 1)<br><br>*Allelic diversity was highest in the Australian region, intermediate in Oceania, and lowest in the Hawaiian Islands (Table 1)<br><br>*Gene diversity was highest in Australian populations (0.849) and lowest on the island of Marquesas (0.393) (Table 1)<br><br>* When looking at the gene loci of Totri 9a it is evident that allelic diversity decreases as you move west to east in the crickets' distribution (Fig 2).<br><br>*On average the Australian region has significantly higher allelic richness and gene diversity than the Oceania and Hawaii regions (Table 4).<br><br>*There is a strong pattern of isolation by distance (Fig 3)<br><br>*Genetic relationships based on microsatellite data suggest the Hawaiian populations are least distant from those in Moorea and the two populations from the Cook | As the crickets move out from their native land of Australia we would expect to see the allelic richness and allelic diversity decrease with each subsequent move to a new island due to the founder's effect. As the distance between the populations increases the amount of gene flow decreases. Because of this it is very unlikely that the crickets colonized the islands through multiple colonization events.<br><br>The movement of the Polynesian settlers matches the spread of the crickets through the islands. It is possible that the settlers helped to spread up this process as the oceans would have been an impediment to the spread of the crickets. |

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| | Islands. (Fig 4)<br><br>*The movement of the crickets is consistent with the models of the movement of the Polynesian settlers (Fig 5) | |
| Bottle necking could contribute to the spread of the flatwing trait | *Bottlenecks were found in only one Australian population, 3 of the 8 populations in the Oceania group, and all three of the Hawaiian Islands. It is also suspected that there is a bottleneck effect in the Marquesas because there is only a single allele at two different loci. (Table 1)<br><br>* On average the Australian region has significantly higher allelic richness and gene diversity than the Oceania and Hawaii regions (Table 4).<br><br>**T. oceanicus* from Hawaii are more likely to mate with a flatwinged male than the females in Australia (Tinghitella & Zuk, 2009)<br><br>*There are low levels of differentiation in Australia and also in Hawaii, Fiji, and the Cook Islands. (Table 2) | The reduction in genetic diversity and expected heterozygosity in the eastern regions as compared with the western regions indicates a recent decrease in population size that is consistent with a bottleneck.<br><br>Other studies suggest that the bottlenecks found outside of Australia may be responsible for the relaxation of female mating requirements. These relaxed mating requirements might have allowed for the spread of the flatwinged trait in Hawaii.<br><br>Low levels of differentiation are expected in Australia because it is a more established population allowing more time for migration to occur. The low level of differentiation in the western islands suggests a high gene flow among these islands allowing for the spread of the non-signaling morph by migrating males. |

Methods

The authors of this paper collected DNA samples from 19 locations in Australia and the Pacific Islands between 2004 and 2007 (Fig 1). These included areas where the crickets have been living for a very long time (ancestral ranges), areas where the crickets and the parasitic fly do not overlap, and areas where the crickets get parasitized by the fly. The leg muscle was

removed the crickets and the DNA was pulled out from this using normal DNA extraction methods. Taking this DNA and using various computer software programs, the authors of the paper were able to look at how similar/different the crickets were to each other at a genetic level. They were looking for things such as how many different genes were present, how many different alleles of those genes were present, and how many of these genes and alleles did the crickets from different areas have in common with each other. From this information genetic trees and global migrations could be estimated using specific software programs.

**Community College Connection**

The topic that this paper connects to in a community college biology course would be that of the Founder's effect and genetic drift. Although natural selection is a powerful player when it comes to changing and forming life on Earth, there are other factors at work as well. Islands are often colonized by a few individuals who then multiply. This is known as the founder effect. Since population sizes are vastly smaller on these island that are "founded" by a few, genetic diversity is low, allowing for a few rare genes to become more prevalent in a population where they might not otherwise (genetic drift).

To teach the concept that there are many selection pressures at work on organisms, some that are random and some that are not, I would create cards that have different scenarios on them for the students to read. Working in a group they would have to decide which kind of selection pressure(s) is at work (ex: natural selection, sexual selection, genetic drift, gene flow). This paper has a nice variety of scenarios that could be used. Examples of some of the scenarios could be: 1. Crickets that "sing" to attract a mate are often parasitized by a fly in Hawaii. Overtime crickets have begun to lose their "singing" ability. 2. Female crickets from one region will only mate with male crickets that sing while female crickets from another region do not discriminate. 3. A small number of crickets get transferred to another island by way of the Polynesian settlers. 4. Australia had the most numbers of alleles present and also contains the oldest populations. 5. Islands that are far away from each other tend to have more genetic differences. Scenarios could be placed on a large chart as a class for a visual example of the different selection pressures. We can talk as a class about how the combination of the selection pressure to be silent from the parasitic fly, and the high level of gene flow between the islands due to people spreading the crickets, has allowed for the quick evolution and spread of the flatwing trait across the islands.

Article #3

**Opposing unfolded-protein-response signals converge on death receptor 5 to control apoptosis**

**Min Lu et al.**

**Problem: What mechanisms control UPR induced apoptotic cell death?**

| Claim/Concept | Evidence/Support | Justification |
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| ER stress induces ligand-independent DR5 activation directly controlled by CHOP. | *siRNA depletion of DR5 ligand APO24L/TRAIL had no impact on the Tg induced apoptosis unlike capsase-8 knockdown (Fig 3A, Fig S3 A-B).<br><br>*Neutralization of extracellular APO24L/TRAIL did not inhibit apoptosis activation (Fig 3B, Fig S3 C-D).<br><br>*DR5 was barely detectable by immunofluorescence in resting SK-MES-1 cells but had an increased abundance with Tg or BfA (Fig 3C)<br><br>*In Tg treated cells DR5 colonized with RACS1 in the Golgi but not the ER marker KDEL, it did however when treated with BfA. (Fig 3C)<br><br>*siRNA depletion of CHOP substantially blocked DR5 mRNA up regulation by Tg or BfA. The knockdown of the CHOP transcriptional targets ERO1a or GADD34 did not. (Fig 4A, Fig S4 A-E) | If this apoptosis pathway was ligand dependent, we would expect to see a decrease in apoptosis when the DR5 ligand (APO24L/TRAIL) was depleted or when APO24L/TRAIL itself is neutralized. This is not the case however suggesting that CHOP has direct control over DR5 activation from within the cell. Further support comes from the fact that a siRNA depletion of CHOP blocks DR5 mRNA up regulation but RRO1a and GADD34 had no effect. It is clear that CHOP plays a direct role in DR5 activation. |
| IRE1α counteracts apoptosis | *siRNA knockdown of IRE1α reduced DR5 mRNA decay in Tg-treated cells (Fig 4B, Fig S4 I-L)<br><br>*A recombinant protein made of | * This suggests that IRE1α might be important in folding. Since IRE1α mediates DR5 RIDD (which |

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| | IRE1α catalytic domains cleaved in vitro transcribed DR5 mRNAs and this was blocked by IRE1α RNase inhibitor 4μ8c (Fig 4C, Fig S4M)<br><br>* CHOP siRNA reduced DR5 up-regulation, caspase 8 activation, and apoptosis. IRE1α depletion augmented these events (Fig 4 D-E, Fig S4 N). XBP1s knockdown lead to IRE1α hyperphosporylation which lead to an increase in DR5 mRNA decay, decrease in caspase 8, and a decrease in apoptosis. 4μ8C enhanced caspase activation by Tg | degrades DNA) it makes sense that we would see a reduction in the breakdown of DR5 mRNA when IRE1α is reduced.<br><br>*This supports the idea that IRE1α has an anti-apoptotic role. IRE1α is essential in regulating the breakdown of DR5 mRNA by RIDD.<br><br>*Together PERK/CHOP and IRE1α work like a teeter totter to keep DR5 in balance and give the cell time to recover from ER stress. Too much DR5 however and that teeter totter gets pushed in favor of apoptosis. |

**Methods**

Different cells types were treated with a variety of ER stress inducing agents. In order to see if DR5 activation was triggered by autocrine death ligand signaling that increases after continual ER stress or if it is controlled directly by CHOP the authors of the paper took some cells and subjected them to something that would either break down Apo2L/Trail (which is the ligand) or caspase-8 (which is needed for apoptosis). They then measured the levels of apoptosis in a control cell and one that had been subjected to ER stressors. They also measured the level of apoptosis when CHOP was depleted and when the transcriptional targets ERP1a and GADD34 were blocked. DR5 activation was measured using immunofluorescence which allows us to take pictures of cells with fluorescent dyes that target specific molecules. The amount of DR5 was also analyzed using QPCR which allows a research to quantify how much of a particular mRNA is present in a sample. Gel electrophoresis (which separates the RNA piece by size) was analyzed to see how the depletion of several factors (like CHOP, XBP1, and IRE1α) affect DR5 and caspase 8

**Community College Connection**

This paper would fit into the section about signal transduction pathways in a community college biology course (specifically the unfolded protein response and apoptosis). The unfolded protein response happens in response to continual stress of the ER which causes unfolded proteins to start to accumulate within the ER. IRE1α can detect these unfolded proteins and starts a series of reactions that turn on genes that help the ER to function better and also to turn on RIDD which degrades DR5 mRNA (which induces apoptosis). All of this gives the cell time to recuperate from this stress. However, having to many unfolded proteins for too long is a dangerous state of being, so as unfolded proteins begin to add up CHOP starts to increase DR5 transcription in the cells. This will lead to apoptosis if enough DR5 accumulate.

To help stress the big idea that everything is in a sort of "balance" within the cell and that small changes can have big consequences down the transduction pathways I would give my students copies of the UPR pathways and the apoptosis pathway (intrinsic and extrinsic). I would give them different scenarios of proteins increasing, decreasing, or being eliminated. They then would have to explain what the result would look like in the cell (apoptosis or not) and why. For example: "What effect would an increase in RIDD have on a cell that is undergoing ER stress?". We could talk as a class about how all these factors are interconnected and work together to help the cell react quickly to its needs.

**Part 2**

Ecology

One of the major "topics" or themes that I took from the ecology course is interdependence. Everything on the planet is dependent on many other factors. No matter what level you are looking at (species, community, ecosystem) nothing stands alone, independent of everything else.

Not only is one particular organism dependent on the resources that are available in its environment but the organism is going to be interacting in some way with the other organisms that share that particular community. Competition is going on between organism vying for the same resources such as food, sunlight, or access to mates. A predator/prey relationship is going to effect the population numbers of both the predator and the prey organism. Some organisms have evolved to have a mutually beneficial relationship (mutualism) such as plants and nitrogen fixing bacteria. Other organisms, like a bird in a tree, benefit from the relationship while the other is not effected (commensalism). In other instances, parasitism occurs when an organism benefits at the detriment of another as seen with mistletoe.

Food webs are an attempt to map out how energy and nutrients moves through a community via interconnected food chains. Change the balance of this food web by, for example, introducing a large number predatory fish to a lake for the sake of fishing, and the ramifications will be felt all along the food chains. A change in the climate might cause a bottom up effect in the food web by decreasing the number of producers in the community. The removal of keystone species (like the sea otter) from a community has devastating effects normally leading to a decrease in biodiversity. Invasive species (often spread by humans) can out compete native species and dramatically alter communities as seen with the Brown Tree Snake in Guam.

Living things are also interdependent on the resources that get recycled through their ecosystems. Phosphorus, for example, is weathered or eroded out of rocks and then is taken up by plants from the water and the soil. Animals can then obtain the phosphorus they need by consuming the plants. The plants and animals will die and decomposers will release that phosphorus again to the soil. Humans are impacting this cycle by adding an overabundance of fertilizers to our crops which run off into the oceans causing eutrophication.

Everything, living and nonliving, is interconnect and dependent on everything else. Humans are included in this web of interdependency. It is vital that we look at the big picture when making decisions that will impact the environment.

Cell Biology

The main topic or "theme" that I took from the cell biology course was regulation. The cell has to have quick and effective regulation so as to be able to adapt to changing circumstances. The cell could not possible obtain enough energy if it had to transcribe a new gene and translate it into protein every time it needed to react to a stimulus, nor would it have enough time.

One of the main ways that a cell regulates its processes is through the use of transcription factors. Only a small amount of a cell's DNA directly codes for transcription factors but these factors can then go on to control the rate of other gene expression by helping or hindering RNA polymerase binding to DNA. Sometimes a transcription factor can even regulate itself by binding to its own gene and serve as a negative feedback. There are often multiple layers of control and often transcriptional factors need co factors to be able to form complexes that RNA polymerase can then bind to. This allows for there to be "backup" pathways in place and keeps the cells from reacting when it is not necessary.

Often there are thresholds that have to be met before a response is seen. In a neuron, for example, voltage gated channels open due to a voltage threshold of around -50mV. If this is not met, then the channels do not open. Ligand gated channels are opened by neuro transmitters which allow Na to enter the cell changing the membrane potential. Enough of the

signal will cause a wave of Na channels opening up along the axon. When it reaches the end of the axon neurotransmitters are released that are premade and stored. All of these interactions insures that the cell only responds when it needs to and that when it does it can do so very quickly. Actin and myosin are a great example of how the cell keeps a ready supply of monomers at hand that it can quickly build from or deconstruct on a moment's notice in order to allow the cell to do things such as move in response to a stimulus.

Posttranslational modification is a great way for the cell to very efficiently regulate proteins. Often this is done through phosphorylation. Kinases and phosphatases can add and remove phosphate groups, making this type of regulation analogous to an on/off switch. Added phosphate groups will alter the protein activity by changing the shape of the protein. This might affect ligand binding by allosteric control or altering the active site. This can also effect the location of the protein as we see this with the nuclear transport receptor. In the case of the ER retention signal KDEL, simply the pH difference between the ER and the Golgi is enough to change the protein confirmation either exposing or hiding the signal.

The cell can also turn off genes through epigenetic techniques such as DNA methylation or creating heterochromatic that wraps up part of a gene so that it is no longer available to polymerases. This allows for some cells to express genes but for others not to. Without this it would be impossible to form a complex multicellular organism.

The cell has many unique ways of regulating its many functions and I only mentioned a few. Like a city, the cell must orchestrate the building of new materials, transportation, and respond to changes as efficiently and quickly as possible.

## Evolution

A main theme that reoccurred throughout the evolution course is that evolution is not directional. There is no goal in mind for evolution and it certainly is not aiming for "perfection". The evolution of organisms is highly dependent on the environment and conditions in which the organism finds itself and often some traits get passed along simply due to chance.

Genetic drift occurs when there are uneven allele frequencies in a population. It is often seen is small populations that are just starting out in an area or have undergone some kind of catastrophe that lowered its numbers. This causes the population to "drift" towards a single phenotype. This phenotype is not necessarily the best for its environment and is selected for simply by chance. We see evidence of genetic drift in the human population. 25% of non-silent mutations are saved in the human population which is much more than other species. This means that natural selection is not getting rid of these mutations and we are carrying around a lot of baggage in or genome that might have been eliminated by natural selection if our population was bigger. We carrying around a lot of deleterious genes. Approximately 1/3 of our DNA is not selectively important. We have a lot of pseudogenes that are not functional but

still present. We also have retrotransposons and DNA that seem to have been inserted by viruses.

This genetic drift is probably the result of the founder effect. We see less and less variation present in our genomes as we move out of Africa. The differences increase with increased distance. As a few individuals branch out and "found" new areas only a small subset of genes are passes on to future generations. We see this same thing happen with the cricket population as they moved out of Australia and into Hawaii allowing for the flatwing trait to become prominent.

People used to feel that the evolution of horses was a great example of progressive change from a small body size to a large body size, but we now know that they really diversified like a tree and that almost all the horse species are now extinct. There was no directional component the large species of horse just happens to be the one that is still alive today.

Resnick's experiments with the guppies showed us that we are able to control for environmental factors and observe evolution happening in a population. There is not a direction in mind it is simply a response from the environment. The change in early humans was spurred on by climate change and the decreasing of the rainforest. Without this change in climate perhaps we would not be here today. Even the crickets in Hawaii today are faced with conflicting pressures in their environment. Natural selection pressure from a parasitic fly to be silent and sexual selection pressure to sing in order to attract a mate.

The evolution of organisms is complex and beautiful, however, it is not direction and there is no end goal. Organisms are either just lucky enough to live to pass on their traits or they are best suited to their environment. Since blind luck and mutations are always a factor we should not expect evolution to arrive at a "perfect ending".

<u>Micro/Immunology</u>

A major topic that I see come up repeatedly during the Microbiology and Immunology course is that of detection and evasion. Our immune system tries to have many different techniques for detecting the invasion of our bodies by pathogens, while the pathogens themselves try to stay one step ahead and evade detection by the immune system.

When a pathogen first enters our cells it is often recognized by components of our innate immune system. Cells like phagocytes can recognize structures on the outside of the pathogen that are not native to eukaryotic cells and then proceeded to phagocytosis them. Those cells can then release cytokines that trigger neutrophils that also use pattern recognition involving the toll receptor family to identify pathogens. The alternative pathway of complement can also be triggered by the presence of a pathogen which will lead to the formation of the membrane attack complex that will rupture the membrane of the invading cell. To avoid the detection of the innate immune system many forms of bacteria have developed tough capsules

that hide the teichoic acids that make them visible to the immune system cells. This helps them to avoid phagocytosis.

Invading pathogens can also come in contact with our adaptive immune system via the lymph. B cells waiting in the lymph nodes can come in contact and engulf the bacteria and then display the antigen to T cells. After activation the B cells can start making antibodies against that particular antigen. These antibodies can activate the classical pathway of complement with leads to a massive amplification response. CD4 and CD8 T cells can travel to the site to destroy pathogens. To combat this bacteria, have a very high rate of evolution. They can easily exchange plasmids with other bacteria and transduction introduces new genes. They are ever changing so antibodies made one day may not be effective the next. Bacteria also produce exotoxins and endo toxins which can lyse host cells or even inhibit protein synthesis.

Viruses tend to hid out inside the host cell but there are still ways of detecting them. When a cell gets infected with a virus it will release interferon which triggers neighboring cells and signals the NK cells to come. The infected cells are either recognized by CD8T cells because they see the antigen presented on MHC1 or they are recognized by NK cells because the cell is not displaying MHC1 (because protein synthesis has been shut down). Extracellular viruses are presented by B cells, macrophages, or dendritic cells via MHC2 molecules to CD4 T cells. Viruses combat this by being highly variable. Replication of viruses is imperfect allowing for the accumulation of mutations. Viral envelope proteins change often (angiogenetic drift) which slows down the response time of the immune system buying them time to replicate and overwhelm the system.

The battle between pathogen and host is one that has been shaped overtime by evolution. Together we will continue to change, always trying to stay one step ahead of the other, and never quite succeeding.