# *UBP* Proposal Example

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## Title: INVADING NEW YORK

Introduction:

*This is an informative and carefully structured introduction: it starts by posing the general research context, briefly stating the specific research goal and justifying its interest. All the information presented is supported by the bibliographic references that are properly quoted.*

New York City is one of the most diverse cities in the world. According to the 2000 Census, 36% of the population of New York City is foreign-born. In addition, there were 48.7 million visitors to New York City in 2010. This large percentage of immigrants and visitors from various countries contributes to the melting pot that is New York City. Everywhere one goes within the city, the evidence of such diversity is apparent. This Urban Barcoding project seeks to investigate what the millions of visitors and residents of New York City bring with them from their native countries. Along with the great diversity of people comes an equally diverse allotment of species, which are shipped here to be sold in ethnic food markets throughout the five boroughs. These different species of fruits, vegetables, herbs, fish and meats are not native to New York or even the United States. Of course, different goods are shipped to New York from countries all over the world every day, but the organisms we will investigate are different in the fact that we hope to find invasive species, which may pose a serious threat to the ecosystem of New York City. These organisms may not only pose a risk to humans but also might pose a threat to the biodiversity of New York City.[1]

An invasive species is defined as an organism that is nonindigenous to a specific location and may be harmful to native organisms by occupying their niches and competing for resources.[2] The question this project seeks to investigate is whether the numerous ethnic food markets in New York City sell invasive species. We will focus on herbs and small plants. We aim to find what possible harm these plants may have on the unique biodiversity of New York City. Even though New York City is such a metropolis, there are tens of thousands of species native to New York. The Department of Environmental Conservation cites the spread of invasive non-native species as the second greatest threat to biodiversity. Since New York City is one of the most frequented gateways into the United States, it is the epicenter of invasive species. In this project we hope to prove this statement is true by finding invasive species being sold throughout New York City.[3]

One example of an invasive species is Blight Fungus (*Cryphonectria parasitica*) which was brought into New York in the beginning of the 20$^{th}$ century on Asian nursery stock. Although already having spent a century in the United States the fungus continues to nearly decimate American chestnut trees and is still a major problem today. Another example is the Northern Snakehead fish (*Channa argus*), native to China, Korea, and Russia. This fish has been found in a pond in Queens, New York. These fish are large predators that out-compete native fish. They are able to survive out of water for

[1] (U.S. Census Bureau 2011)
[2] (Bustamante, C, & Taylor, J., 2011)
[3] (Department of Environmental Conservatism 2011)

several days in damp areas and can even move across ground to gain access to water. This species is very dangerous to native aquatic life since they feed on fish, frogs, and aquatic insects and can live in virtually any lake or river. Lastly, the Asian long horned beetle (*Anoplophora glabripennis*) is noted as the one of the most destructive invasive species. It is capable of destroying native species of trees. This species was also introduced to the United States through New York, brought in on shipping crates. Invasive species are such a major issue that President Clinton issued Executive Order 13112 on Invasive Species. This order created an Invasive Species Consul in order to monitor invasive species in the United States and prevent further invasive species from being introduced.[4]

Invasive species usually first appear in areas where the people living there and the species have a common ethnic background. For this reason we will investigate ethnic food markets since there are so many ethnicities that call New York home. New York City is wonderfully diverse; however, this project's questions whether the plethora of non-native organisms sold here may be potentially harmful to the biodiversity of New York City.

Methods:

*Because all UBP teams will use common overall methodology (DNA extraction, PCR and Sequence Analysis) that is clearly described in the protocol provided (www.urbanbarcodeproject.org/files/Barcoding_Protocol.pdf) the most important details to include here are your sample collection/storage procedures.*

New York City is a much diversified area of backgrounds and cultures; this corresponds to our group itself, because just as a multitude of cultures has gathered together as a city, our group is also a mix of heritages. It is a representation of the diversity that exists here. As a way to incorporate our nationalities into the project, we will look into each team member's cultural background to find associated food markets that have possible sample materials. In this way, we will be able to collect our data samples from different ethnic markets around the city, most likely in those specific ethnically dominated areas, to discover the varied species of common remedies used in our cultures.

Our team will take day trips to culturally distinct areas of New York City to find an ethnic market associated with the four differing nationalities our team possesses. We will take two trips to each to make sure the contents of our purchases are consistent after time elapses. Our plan is to find specimens not normally available for purchase in our everyday supermarkets, but still very much a part of the culture of New York City. We are hoping to focus on herbs and small plants sold in cultural markets. Our plan is to learn the uses for the herbs we collect and, once we sequence them, compare them to herbs more commonly used in this country. We may find close taxonomic connections between the samples we collect and genetic sequences of herbs commonly found in supermarkets. By doing so we might discover new uses for herbs we find in our supermarkets by learning about herbs from ethnic food markets.

Upon collecting our samples, we will accurately document each sample itself along with the locations in which we bought them. We will add all data to the sample database (https://sampledb.dnalc.org). This includes picture documentation, of both the locations where the specimen was obtained as well as pictures of the samples, and accurately labeling of all the samples after putting them into separate tubes. We will take proper measures to preserve each of the samples, most likely by means of refrigeration, where each sample will be stored in a separate freezer bag with the sample's name and location where the sample was procured.

Our group will bring all collected samples to the DNALC laboratory in Harlem so we can prepare our samples for analysis. We will cut small samples, 10–20 mg, from each of our specimens and

[4] (Clinton, 1999) ("Conservative Issues: invasive,") (Griffin 2010) (Bustamante & Taylor, 2011)

contain them in different, labeled centrifugation tubes. We will also take precaution to avoid cross-contamination by cleaning scissors and tweezers with alcohol between samples. We will add 300 µL of lysis solution to each tube, and forcefully crush each of the samples with a pestle for at least two minutes. We will heat the tubes at 65 degrees Celsius for ten minutes. We will then place the tubes in a balanced centrifuge, with the cap hinges pointed upward, and centrifuge at maximum speed for one minute. We will transfer 150 µL of the supernatant to a fresh tube.

In order to separate nucleic acids from the extraction solution, 3 µL of silica resin will be added to the tube and mixed well by pipetting up and down. The tube will then be incubated at 57 degrees Celsius for five minutes. Afterward, the tubes will be centrifuged for 30 seconds at maximum speed. After pouring out the supernatant, we will add 500 µL of wash butter to the pellet and resuspend the silica by pipetting up and down. This step will remove contaminants from the sample while the DNA will remain bound to the silica. We will then centrifuge the tube at maximum speed for 30 seconds. Once again, we will pour out the supernatant and add 500 µL of wash buffer to the pellet and resuspend the silica by pipetting up and down. The tubes will then be placed in a balanced centrifuge for 30 seconds. The supernatant will be poured out, and the remaining supernatant will be removed using a micropipette with a fresh tip.

In order to elute the DNA from the silica, we will add 100 µL of distilled water to the pellet and mix well by pipetting. The tube will be incubated at 57 degrees Celsius for five minutes. We will then centrifuge the tubes for 30 seconds to pellet the silica resin. Since our DNA will now be in the supernatant, we will transfer 50 µL of the supernatant to a fresh tube while being very careful not to disturb the pellet while transferring the supernatant.

We will store the tubes on ice until we are ready to pipette 2µL from each sample, which we will put into the PCR tubes. Once this is completed, we will mix the samples with loading dye and load them into a gel electrophoresis. We will run the gel and compare the samples to ensure proper DNA isolation. We will then send off the samples to be sequenced, which will then be organized and compared to sequences found of the DNAsubway.org website.

Once all of the samples have been analyzed, we will be able to identify the individual ingredients and seek the legality of their existence here in this country. We will compare the actual makeup of the supplements to the listed ingredients to find any inconsistencies and possible reasons as to why they may not list those ingredients. Upon further examination, we hope to determine some possible effects the material can have by searching them out on the New York Invasive Species Website. In we way, we can seek out any danger it may pose to the biodiversity of New York City.

Specific Aims:

*This section provides a clear statement of the research project specific goal, a logical and consistent estimation of expected results and the overall impact of the project. All the information is backed by a referenced bibliography properly quoted throughout the text.*

In this Urban Barcoding Project, we seek to investigate the numerous ethnic food markets in New York City. In these markets we will search for herbs and small plants that might be invasive species. We hypothesize that we will find many examples of invasive species throughout our investigation. We have formed this assumption because of the numerous examples of invasive species that we have uncovered in our research of the invasive species of New York as cited by New York Invasive Species Information. The number of invasive species in New York is so great that a full list has never been compiled. We

especially think it will be fruitful to focus on small plants and herbs because there are more than 50 species of plants named as invasive to the biodiversity of New York.[5]

One example of an invasive plant is Honeysuckle. Honeysuckle was first brought into America in the 18th century. Today it can be purchased online. The plant is from Southern Russia and Asia and readily out-competes native Honeysuckle. Honeysuckle can also have negative effects on birds because its berries have less nutrition than the berries of the native Honeysuckle. By researching invasive plants such as these we have learned that there are an overwhelming number of invasive and potentially invasive species. We have also learned that these invasive species do not only out-compete native species but also have an impact on an entire ecosystem. We aim to uncover if we can find any invasive species being sold in ethnic food markets. Once we have discovered if they are invasive or not, we also hope to discover what impact the species can potentially have on the biodiversity of New York City.[6]

Our group aims to inspect the foods available in the many ethnic food markets of New York City. These markets are often the gateways of invasive species, since the items may be imported secretly or without any restrictions. This has potential hazardous implications, as these non-native organisms can compete for the niches occupied by the native organisms of New York City. As evinced from the plethora of examples mentioned of invasive species found in New York City, we believe that many organisms sold in these markets are in fact deleterious to our environment, specifically by posing a great threat to the biodiversity of New York City. These organisms, especially plants, easily disperse seeds or take root which pose an immediate threat to native species. Since these species have no natural predators they multiply quickly and are unnoticed until they take over the niches of New York City's native species.

In our analysis, we expect to discover species that are illegal to our country, and possibly are being sold under the guise of a native organism. These species may be overlooked due to intentional mislabeling. New York City is described as the melting pot, meaning it is incredibly ethnically diverse. Our goal is to explore various species available for purchase from different ethnic & cultural food markets around New York City and its native inhabitants to reveal something that may potentially be injurious to our distinct city.

Data Analysis. *The students should include a brief explanation on how they plan to analyze and interpret the results obtained.*

| Species | Date Purchased | Location of purchase | Organismal DNA not originally indicated | Organismal DNA identified | References in database |
|---|---|---|---|---|---|
| Species 1 sample a | | | | | |
| Species 1 sample b | | | | | |
| Species 1 sample c | | | | | |
| Species 1 sample d | | | | | |

[5] (NY Department of Environmental Conservatism, Biodiversity and Species Conservation, 2011)
[6] (*Honeysuckle*, 2011)

| | | | | | |
|---|---|---|---|---|---|
| Species 2 sample a | | | | | |
| Species 2 sample b | | | | | |
| Species 2 sample c | | | | | |
| Species 2 sample d | | | | | |

We plan to collect at least three samples of each species, or what we believe to be the same species, as we want to ensure the authenticity of the outcome of our results. We anticipate that we will focus on herbs and small plants. New York City is known as the melting pot, it is important for us to have a sense of the location where the specimens were obtained, such as Chinatown, Greenpoint, and Little Italy that is why we will keep careful documentation of where the specimens were purchased. We want to keep the data well organized because many labels are in different languages. We will translate these labels and then further investigate whether these products are true to the ingredients that are listed. We plan to compare the organisms indicated on the sign or ingredient label to the organism we find in our DNA analysis. We anticipate some discrepancies. Once the species is known, we will compare our finding to the database of invasive species of New York.

References:

*This is a substantial and varied list of references. For key bibliographic resources on barcoding please refer to:* http://www.urbanbarcodeproject.org/resources.html


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