The Search for Leonardo's Genome

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In the end, my argument is that the search for the genome of Leonardo Da Vinci reveals many potential benefits of a more frequent marriage of new biology, in particular genomics and microbiology, with art, art history, and the conservation of cultural heritage. To begin, consider a trio of entertaining studies of the scandalous power of human genomics. In a large survey of parenthood conducted in the United Kingdom, in which parenthood was not in dispute, one in twenty-five fathers was not the biological parent. (Ref 1) In a similar survey in which parenthood was disputed, 30 percent of the fathers were not the biological parents. In a 2019 study conducted at the U.S.-Mexico border, 30 percent of those tested were genetically unrelated to the children they claimed as their own. (Ref 2) Biology can both end and begin mysteries.

Now consider the power of art as demonstrated by money. The global art market in 2021 was valued at approximately $65 billion. About 40 percent went through New York City, with Hong Kong the second art market capital, followed by London, Paris, and Geneva. Most of the demand is for postwar, contemporary, and modern art. Sales of Old Masters, such as Leonardo and others who worked in Europe before 1800, make up less than 5 percent.

Attribution and authentication are crucial matters for buyers, sellers, and intermediaries, including dealers and auction houses. Hundreds of years ago the art market invented blockchain, a fancy word for reliable provenance. Provenance and connoisseurship—intelligence without artificiality—establish identity in art markets.

Identity requires comparison – a reference and hopefully a match. The match can pair fingerprints, retinas, voices, faces, or other images or attributes. Natural history museums and botanical gardens preserve a so-called holotype, a single physical example of a reliably described organism, in a jar or drawer as a reference specimen against which to establish the identity of other specimens. Taxonomists build botany and zoology on such collections of plants and animals.

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Mentioning the description of animal species offers me a chance to explain a thread in my own story. I have devoted much of my career to censusing the diversity of marine life. Until recently, to identify most marine animals one had to capture them, which can be harmful, or photograph them extensively, which is often difficult in a vast dark ocean. Over the past twenty-five years I have been part of a community that has developed affordable, scalable techniques for identification that are much less harmful to the organisms, such as collecting the DNA that the animals shed in sea water and recording the sounds the animals make.

Short "DNA barcodes" of as few as one hundred letters representing the four bases (cytosine [C], guanine [G], adenine [A] or thymine [T]) that make up a DNA strand, a tiny fraction of the genome, usually suffice to identify the species of a fish from the Hudson River or the middle of the Atlantic Ocean. (Ref 3) Of course, a carefully documented specimen properly preserved in a jar in a museum provided some tissue whose DNA offers the sequence, deposited in a reliable database, against which an investigator makes the match. In addition to DNA, other molecules can accomplish these identifications. So too can the collection or profile of microbial organisms that live on a critter or inside its cheek, what microbiologists call a microbiome. These microbiomes are ubiquitous; our world is not sterile. Microbes abound in a carpet, on a piece of wood, and on the surface of a sheet of canvas or paper.

The art world, exemplified by the extraordinary research labs of the Metropolitan Museum of Art in New York, the Smithsonian Institution in Washington, D.C., the Getty Center in Los Angeles, and their counterparts in England, France, Italy, and other countries, has been utilizing chemistry and physics for many decades. X-rays and other types of radiation reveal images; carbon-14 and other radioactive isotopes disclose dates; and mass spectrometry and gas chromatography identify materials used in the art. In cultural heritage, biology has tended to be treated like dirt or contamination to be cleaned away. My suggestion, to use a phrase popular in Silicon Valley, is to treat biology as a feature and not as a bug.

Let me share another personal thread. Why and how did I become interested in Leonardo? Among my most important mentors is the Italian physicist, Cesare Marchetti, who recently turned ninety-five. About twenty-five years ago, during a visit to Marchetti's home in Tuscany, he asked if I had studied Leonardo, to which I answered no. His response was that the English read Shakespeare, the Germans Goethe, and the Russians Tolstoy, and a good education in Italy must include Leonardo. I should not travel through life without some exposure to Leonardo. He gave me a copy of a book from the 1938 Milan exhibition, republished in 1996, about Leonardo's achievements, spanning astronomy to zoology. (Ref 4)

Over the next fifteen years or so, we had endless fun with Leonardo's puzzles and lists. I became interested in hidden images, anamorphoses, which Leonardo

played with here and there. (Ref 5, Ref 6) We called them cryptos. A magnificent painting in the Metropolitan Museum by Leonardo's master Verrocchio has a hidden image of the head of a kite (nibbio), a bird featured in Leonardo's drawings and earliest memories.[2] Perhaps Leonardo snuck the image into the painting. His tiny or micro images are also intriguing. How could anyone draw so precisely and accurately at the scales at which Leonardo worked?

Jump ahead to the year 2014. Marchetti and I are conversing with Brunetto Chiarelli, a bone-and-tooth expert who was then the head of the Institute of Physical Anthropology at the University of Florence. Chiarelli suggested we establish a project to obtain and sequence Leonardo's genome. Chiarelli believed that with the collaboration of his colleague Henry de Lumley, head of the Institute of Human Paleontology in Paris, a research team might gain access to the tomb of Leonardo in Amboise in the Loire Valley at the Royal Chateau, where Leonardo spent the last three years of his life under the patronage of Francis I, King of France. De Lumley was keen to join our effort, and so the Leonardo Da Vinci DNA Project began.

De Lumley cautioned that the Count of Paris, who as head of the Fondation St-Louis controls the Chateau, was unlikely to let us disturb the beautiful tomb, except possibly as a capstone to the entire project if much of what we envisioned doing proved successful. We would need undeniable reference materials, like holotypes, against which to compare the materials that may or may not be in the tomb. By the way, the tomb was moved and opened during the French Revolution and again later in the nineteenth century, and perhaps on other occasions too. (Ref 7) In any case, the animating question for the Project became, do the relics that lie in the Amboise tomb contain Leonardo's DNA?

Our list of questions quickly grew and included a few about Leonardo's ancestry, in particular about his mother. We know much about the family of Leonardo's father, a consequential notary, but distinguished Leonardisti, such as Martin Kemp and Alessandro Vezzosi, hold widely divergent views about his mother, who was probably a local Tuscan peasant or, more dramatically, an enslaved person brought from the Middle East or from Circassia north of the Black Sea. (Ref 8, Ref 9) Did Leonardo have an unusual parental combination?

A third set of questions, which makes the project appropriate for a biomedical research institution like The Rockefeller University, refers to Leonardo's extraordinary visual acuity in both space and time. Project member David Thaler (University of Basel) has published a remarkable pair of papers exploring evidence for Leonardo's exceptional vision. (Ref 10; Ref 11) Could Leonardo have excelled in the present as

[2] https://www.metmuseum.org/art/collection/search/437892 The forefinger and middle finger of the left hand of the Madonna are inside the head of a white bird. A bird also pecks at the right nipple of the child.

a baseball batter or a soccer goalie? Famous art historians and biographers such as Sir Kenneth Clark and Walter Isaacson have written about Leonardo's "quick eye" because of the way he accurately captured fleeting expressions, wings during bird flight, and patterns in swirling water. Until now no one had tried to put a number on this aspect of Leonardo's extraordinary visual acuity. Thaler notes that flicker fusion frequency (FFF)—akin to a motion picture's frames per second—is used to quantify and measure "temporal acuity" in human vision. When frames per second exceed the number of frames the viewer can perceive individually, the brain constructs the illusion of continuous movement. An average person's FFF is between 20 to 40 frames per second; motion pictures today present 48 or 72 frames per second. To see accurately the angle between dragonfly wings would require temporal acuity in the range of 50 to 100 frames per second. Further study could compare the genome of individuals like Leonardo and species with unusually high FFF.

A fourth set of questions relates to Leonardo's diet and health. From verified bones, we may be able to learn whether he maintained a vegetarian diet for much of his life, as is widely believed.

A fifth question, or possibility, is to reconstruct Leonardo's appearance, that is, to go from genotype to phenotype. With enough of the genome, one can now do a reasonable job of reproducing physiognomy. Team members from Craig Venter's Institute have pioneered a technique using genomes to predict what faces look like. (Ref 12) Carmen Bambach, curator of drawings and prints at the Metropolitan Museum and the author of a monumental four-volume study of Leonardo, neatly summarizes a long-standing debate about representations of Leonardo, including the famous image in Turin that may or may not be a self-portrait. (Ref 13)

In the same spirit, team members Francesco Galassi and Elena Varotto of Sicily's Forensic Anthropology, Paleopathology, and Bioarcheology Research Center analyzed shoes that belonged to Michelangelo Buonarroti to estimate his height at 157 centimeters (5 feet 2 inches). (Ref 14)

How do we set off on this journey that might conclude at Leonardo's tomb in Amboise? An obvious route is via possible living descendants. Leonardo was gay and no mention has ever been found that he sired children. While he was the only (and illegitimate) offspring of his father and mother, his father sired seventeen other children by several wives. In 2021, under the leadership of historians Alessandro Vezzosi and Agnese Sabato, the Project published "The New Genealogical Tree of the Da Vinci Family for Leonardo's DNA: Ancestors and descendants in direct male line down to the present XXI generation," an open access ninety-page booklet with a 690-year genealogy that identifies fourteen possible living descendants. (Ref 15) Bastards, of course, could intervene, but during April 2022 six of the fourteen possible descendants gave their saliva to David Caramelli's Laboratory of Anthropology, Molecular Anthropology, and Paleogenetics at the University of Florence in order to

study their Y chromosomes, which are passed largely unchanged from father to son.

One aim of the project is to match the DNA of the descendants against materials found in the tombs of descendants in the Vinci area from say 1600, tombs that for artistic reasons are unremarkable. Eventually we might seek to open the tomb of Leonardo's father, Ser Piero, and half-brothers in the beautiful Badia Fiorentina near the Palazzo Vecchio in central Florence. Renaissance historian Anne Leader has identified the likely spot of the tombs (Ref 16), and experts have conducted studies with ground-penetrating radar to try to localize the bones (Ref 17). But opening an artful historic tomb is a serious matter in Florence as it is in Amboise, and the Florence floods of 1966 may have damaged the contents.

The most intriguing possibility is to obtain DNA off the pages of the notebooks or off drawings, in particular drawings made by metal or silver point, in which saliva is used to prepare the paper, a possibility introduced and explored by artist Karina Aberg. The laboratory of Thomas Sakmar (The Rockefeller University) has been working with great success on techniques to obtain DNA off papers of diverse kinds and ages. The late scholar and dealer Fred Kline provided the Project with fourteen works on paper with some provenance and/or authentication that are being used in our experiments. Colleagues in Spain, Jose Lorente and Christian Galvez, who are interested in Columbus and other historical figures are also advancing the techniques, and exploring the possibility to obtain samples from the six hundred pages of Leonardo's notebooks held by the Spanish National Library in Madrid, some of which appear to have Leonardo's fingerprints on them. (Ref 18, Ref 19, Ref 20)

While writers and artists tend to handle the edges or borders of sheets of paper, leaving DNA there, greater historical and artistic value typically and fortunately may be found in text and images that are more centrally located. In any case, contamination from many people handling the pages of the notebooks or other sheets as well as other works, such as paintings, is a major concern. The project's lead contamination expert, Rhonda Roby, now at California's Alameda County Sheriff's Office, has extensive forensic experience solving famously difficult cases. (Ref 21)

Finally, other relics, such as hair or a ring, might merit examination. Unsurprisingly, collectors often contact us about objects they hope to associate with Leonardo, for example, a globe made from an ostrich egg that depicts what Europeans called the New World. (Ref 22)

In any case, the key to the game is a match of two or more sources, such as a living descendant's Y-chromosome DNA with a comparable sequence from a notebook page. Once we have that key, we hope to grow the reliable sequence and search systematically, for example, for the genes that influence vision.

While Leonardo presents particularly intriguing challenges, the ideas of the project are now "in the air." In 2014, the English were excited to find, with a 99 percent probability, the bones of Richard III in a parking lot in Leicester. (Ref 23)

The techniques for locating and piecing together ancient DNA are getting better. Caramelli's lab works on Neanderthals from thirty thousand years ago and armies that clashed in Sicily in 480 BCE. (Ref 24) In another fifteen to twenty years, much of the genomics and microbiology we are doing will become customary parts of history and conservation sciences.

Whether or not we succeed with Leonardo's genome, a movement to integrate scholarship in biology and art is growing, led by Julie Arslanoglu of the Metropolitan Museum of Art and Margaret Holben Ellis of NYU's Institute of Fine Arts. They have hosted two conferences on "Art Bio Matters" and have started a community website.[3]

Let me end with a few general comments. One dimension that we did not anticipate and that elicits great interest concerns fraud and forgery. Artists such as Dali, Rothko, and Basquiat are bedeviled by these problems. Building a database of forgers' DNA and including the DNA of artists could help. Living artists might want to deposit their sequences to lessen the chance of future fraud. Project lawyer Eric Rayman has helped raise the salience of DNA and art law. (Ref 25)

A second growing dimension is degradation and microbiomes, previously mentioned. Team members Manolito Torralba and Karen Nelson (formerly at the J. Craig Venter Institute in La Jolla, California) and other colleagues used small, dry polyester swabs to gently collect microbes from centuries-old, Renaissance-style art in a private collector's home in Tuscany. Their findings published in the journal *Microbial Ecology* show that much remains to be learned about how to slow or reduce degradation and also about how to preserve works in conditions of changing air chemistry and climate, not only in museums but elsewhere. (Ref 26)

I also mentioned that biology is a feature and not a bug. We need to ask whether we have been cleaning too aggressively, or without proper preservation of what is removed. New York City's Morgan Library is not only a library but a biological repository that may have the DNA of Mozart, Thoreau, Gertrude Stein, and Sylvia Plath. Appreciating genetics and microbiology might increase the value of many collections.

In this vein, we may learn a lot by studying extraordinary human performance, perhaps outliers, of several kinds. The New York Public Library for the Performing Arts has begun a project with biologist Andrew Miranker at Yale University to try to obtain the DNA of Franz Liszt from materials that belonged to him. Liszt experienced synesthesia; he saw musical notes and chords as colors. DNA might also give us wonderful clues to the anonymous artists who worked in ancient Egypt or medieval Ireland or the Kingdom of Benin.

Finally, while the Leonardo DNA Project is decidedly nonprofit and publishes

[3] Art Bio Matters, https://www.artbiomatters.org.

its findings open access, a lucrative entrepreneurial opportunity exists to provide services to auction houses, galleries, and collectors, most powerfully by an enterprise that encompasses genomics, artificial intelligence, and expertise. For example, scanning the more than seven hundred works attributed to Vincent van Gogh would allow a machine to learn deeply about van Gogh, while his DNA could be obtained from his clothing and other personal items in the collections held by the Van Gogh Museum in Amsterdam. I hope I have persuaded readers that such marriages, exemplified by the search for Leonardo's genome, could overcome formerly daunting limits to knowledge and explore not only the unknown but what seemed unknowable.

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