Matrilineal Lines:
Mitochondrial DNA Tests

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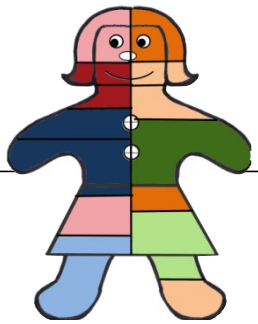
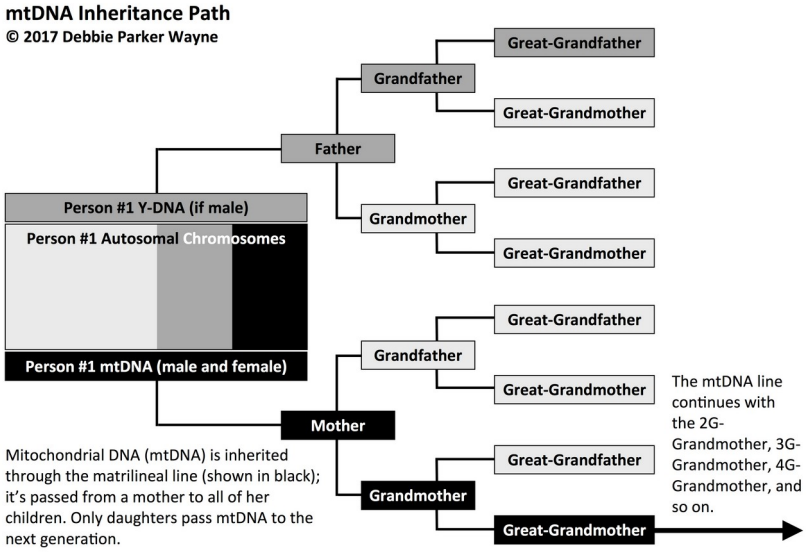


Mitochondrial DNA (mtDNA) may be the least used type of DNA, but is the only type of DNA test that can answer some genealogical questions. mtDNA is inherited from your mother who inherited it from her mother who inherited it from her mother and so on back in time. Because mtDNA does not recombine as it is passed down to the next generation it can trace this one line—the matrilineal line—back hundreds, even thousands of years. Both men and women inherit mtDNA and can take an mtDNA test. But the only line represented in an mtDNA test is the matrilineal line as shown in figure 1 (mtDNA Inheritance Path).



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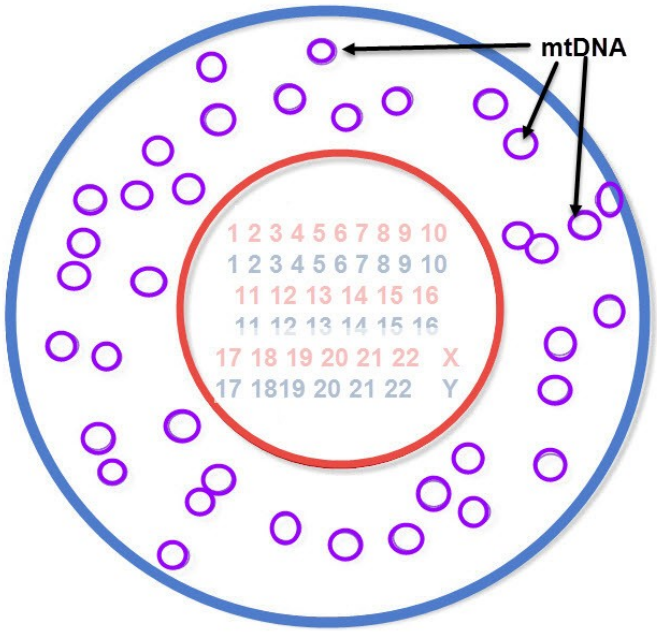
WHAT IS mtDNA?

Mitochondrial DNA is separate from the DNA in our chromosomes and is found outside of the cell nucleus. A cell of our body is illustrated in figure 2 (mtDNA in the Cell) with arrows pointing to some of the hundreds of mitochondria that are found in every cell of our bodies.



mtDNA in the Cell

One mitochondrial molecule is depicted in figure 3 (mtDNA Molecule). It is a small circular segment of DNA consisting of 16,569 locations (also called base pairs). The exact number of locations will vary if there are insertions or deletions (mutations where base pairs are added or lost). The segments on either side of the start of the circle change or mutate more often than the rest of the molecule. These are called hyper-variable regions or segments (HVR, HVS). The exact start and stop points of hyper-variable regions vary between testing companies even when the same name is used; the numbers shown are used by Family Tree DNA and some other companies. The rest of the mtDNA molecule is the coding region and may contain some medically-relevant information.

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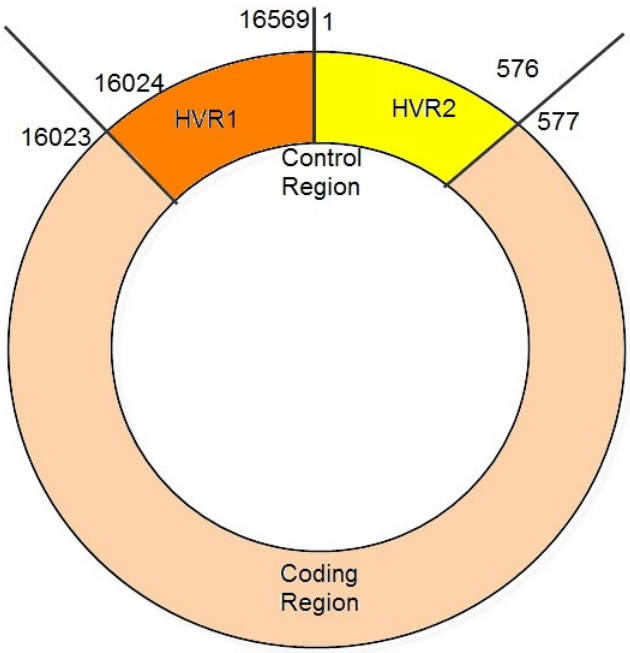

mtDNA Molecule

mtDNA INHERITANCE

Mitochondrial DNA is passed from a mother to all of her children. Daughters pass it to the next generation. This inheritance pattern is illustrated in figure 4 (mtDNA Inheritance) where men are depicted as squares, women as circles, and pink indicates the mtDNA inheritance path from the woman on the top row. Green, yellow, and blue indicate mtDNA passed from women who marry into this family.

The mother on the top row (person number 2) passes her mtDNA to her son (3) and daughters (6, 8). The son (3) does not pass his mtDNA to his children. Daughter (6) passes her mtDNA to her son (13), but the son does not pass his mtDNA to his children. Daughter (8) passes her mtDNA to her daughter (16) who passes the mtDNA to her son (21) and daughter (22). Of the descendants shown on the bottom row, only daughter (22) will pass the mtDNA of matrilineal great-grandmother (2) to the next generation.

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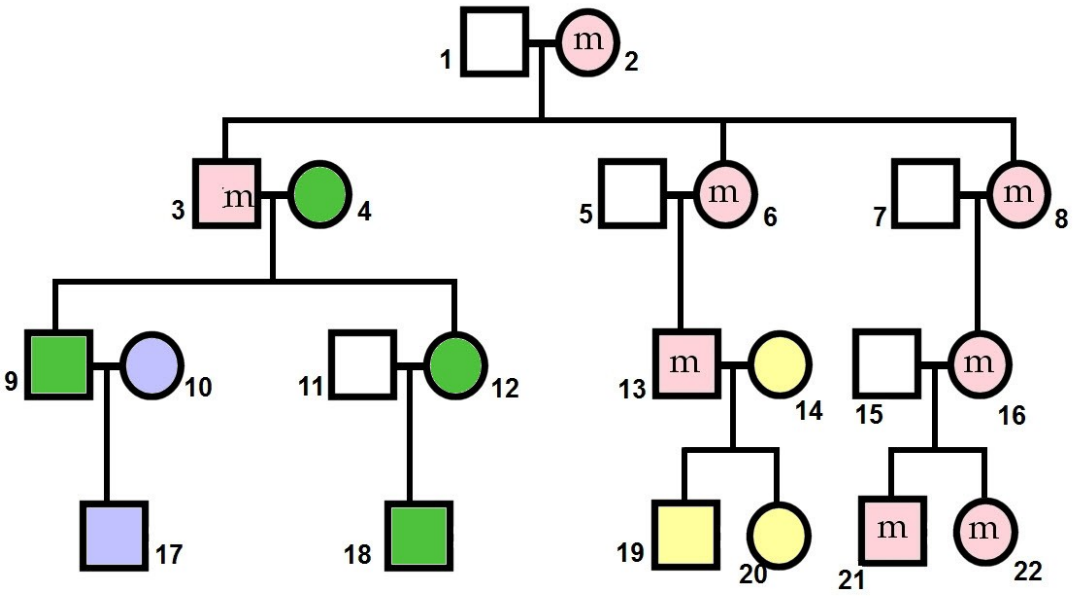

mtDNA inheritance © 2009, Debbie Parker Wayne

The mtDNA passes from mother to child unchanged, unless a mutation occurs. A mutation is a change caused by a copying error during the DNA duplication process. Mutations in mtDNA rarely occur, but do occur at random intervals and locations. These mutations allow us to trace a family tree using DNA, grouping those with like changes.

mtDNA TESTS

Family Tree DNA (http://familytreedna.com/) offers two mtDNA tests. The mtDNAPlus test sequences only HVR1 and HVR2. The mtFullSequence test sequences the entire mitochondrial molecule. While the autosomal DNA tests offered by other companies look at a small number of mtDNA base pairs, those companies do not provide mtDNA databases that allow

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comparison of the mtDNA results to other test-takers. Other companies offer mtDNA tests, but Family Tree DNA has a large database and generally offers the best prices with frequent sale discounts.

Today the full mitochondrial sequence (mtFullSequence) test is affordable for most genealogists. It is the most useful for genealogical research, but test results should be analyzed for medical relevance, if any, before the results are disclosed to others. Because relatives have the same DNA, revealing information about your DNA could also reveal information about your descendants, ancestors, and collateral relatives. An informed decision about sharing data can be made only if you know what might be revealed.

> Note: A Full Mitochondrial Sequence (FMS) should not be confused with a Full Genome Sequence (FGS) or Whole Genome Sequence (WGS). The phrase Full Genome Sequence or Whole Genome Sequence refers to the sequencing of a person's entire complement of DNA (autosomal, X, Y, and mitochondrial DNA), not just all of the mitochondrial DNA.

mtDNA TEST RESULTS

Family Tree DNA publishes statements on the likelihood of a common matrilineal ancestor being found in a specific number of generations as shown in table 1. Even when the full mitochondrial sequence is an exact match there is still a 50% chance your common ancestor is more than five generations back. For this reason a more complete matrilineal lineage makes it more likely an mtDNA connection can be identified. Even with these low percentages of the common ancestor being in recent generations, mtDNA can solve many genealogical problems.

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Table 1. mtDNA Regions and Common Ancestor Matches			
Segment name	HVR1	HVR2	Full mtDNA Sequence (FMS) HVR1, HVR2, and coding region (CR: 575 to 16,000)
Locations tested	16,001 to 16,569	1 to 574	1 to 16,569
Common ancestor computations	50% chance there is a common ancestor within 52 generations (about 1300 years) for two people with exact matches	50% chance have a common ancestor within 28 generations (about 700 years) for two people with exact matches	50% chance you have a common ancestor within 5 generations (about 125 years) for two people with exact matches[a]
a. " What are the parts of the mitochondrial DNA (mtDNA)? What are HVR1 and HVR2? What is the Coding Region?," The Family Tree DNA Learning Center BETA, *Family Tree DNA* (https://www.familytreedna.com/learn/mtdna-testing/parts-mitochondrial-dna-mtdna-hvr1-hvr2-coding-region/). "How many generations back does mitochondrial DNA (mtDNA) testing trace?," The Family Tree DNA Learning Center BETA, *Family Tree DNA* (https://www.familytreedna.com/learn/mtdna-testing/generations-traced/).			

Mitochondrial DNA test results consist of several parts.

1. A match list of others in the company database whose mtDNA closely matches yours, as shown in figure 5 (mtDNA - Matches). To see all close mtDNA matches be sure the filter is set to show matches for "The Entire Database." If there are no close matches for all regions (HVR1, HVR2, Coding Regions) the display will drop back to show matches only on the HVR regions.

   Genetic distance, the first column, indicates how many locations differ between you and the matching person listed in column 2, Name (partial names are blurred here for privacy reasons). A genetic distance of 0 if a full match, a genetic distance of 1 is a one-step difference or mismatch, and so on.

   The third column has clickable icons allowing email contact and saving notes regarding your match to this person, an icon indicating whether the person has posted a family tree, and a list of tests taken by this person. The fourth column lists the most distant ancestor if the test-taker has entered one. For the matrilineal line this should be the most distant ancestor in the mtDNA line, but some test-takers enter

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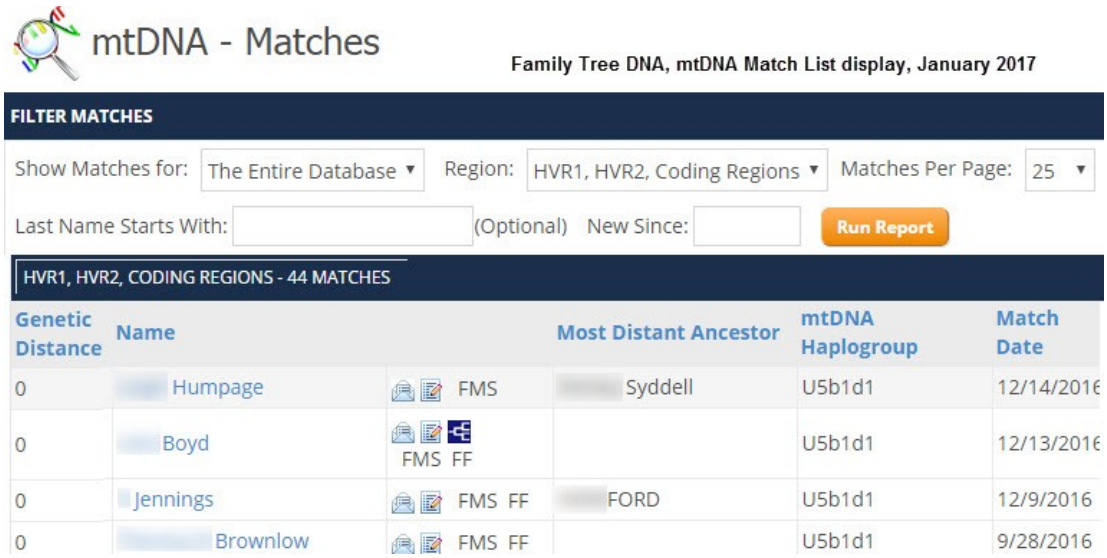
other ancestral lines here so be sure to contact the test-taker to confirm the lineage. The haplogroup and match date are in the last two columns.



2. The haplogroup will be identified as, for example, W6c, U5b1d1, or X2a1a. This represents the deep roots of the matrilineal ancestry—the location of ancestors tens of thousands of years ago. Two people in the same haplogroup share a common ancestor, but it might be thousands of years ago. A full mtDNA sequence usually results in a more refined haplogroup assignment. A test-taker who takes only the mtDNAPlus test may be identified as haplogroup U5 while the same person taking the mtFullSequence test may be identified as U5b1d1. Haplogroup assignments may change over time as the mtDNA phylogenetic tree is refined due to advancing DNA research.

3. The location and chemical value of the mtDNA **differences** (or mutations) between a reference sequence and the person tested. The chemicals are Adenine, Cytosine, Guanine, Thymine, each usually represented by the first letter of the name—A, C, G, or T. This list of differences should be compared to the test results of others to find potential relatives. The only information you will have is the genetic distance unless you contact the matching person and ask them to share the list of differences. As shown in table 1, even with an exact

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match on a full mtDNA sequence there is a chance the common ancestor is many generations back in time.

There are two reference sequences in use. These are the Cambridge Reference Sequence (CRS) and the Reconstructed Sapiens Reference Sequence (RSRS). When comparing test results always be sure both sets of results were compared to the same reference sequence. You cannot compare your RSRS results to someone else's CRS results. Click on the tab for the reference sequence to toggle between the two on the mtDNA results page.

The mtDNA test results will be a list of values similar to those shown in figure 6 (mtDNA – Results, this example uses the RSRS sequence).

- Numbers represent the location on the mtDNA molecule.
- The first letter indicates the chemical that would be found in an ancient ancestral sample at the numbered location. This value is implied in a CRS comparison and not explicitly stated.
- The second letter indicates the chemical found in the tested sample at the numbered location.
- A deletion would be indicated by a "D" (not seen in this display).
- Additions are indicated by a period followed by a number and a letter representing the chemical at the location. The list indicates an added Adenine (A) and Cytosine (C) base following location 522.
- "Extra Mutations" are mutations that are not plotted on the phylogenetic tree (the human mtDNA tree). Some of these are common mutations found in multiple branches of the tree; some are recent mutations that may provide evidence of a close link between two test-takers.

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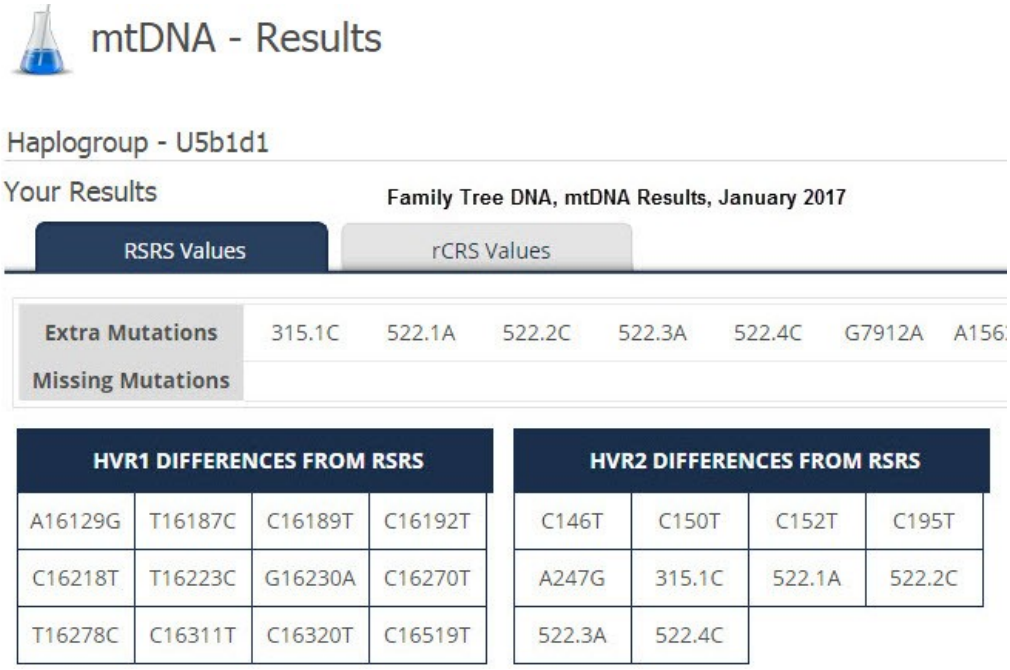

An mtDNA exact match will have the same values at the same locations. A one-step difference (genetic distance 1) would have a different chemical at one location, a two-step difference would have a different chemical at two locations, and so on. The more differences there are the further back in time the common ancestor is likely to be found.

4. If the mtFullSequence test is taken, the test-taker can download a file containing the "raw data" or the chemical values at each location. This file is in a format known as FASTA. Scroll to the bottom of the mtDNA Results page to find a button that allows the FASTA file to be downloaded to your personal computing device. The file consists of a kit number (>12345), identification of the regions tested (HVR2,CR,HVR1), and row after row of ACGT representing the 16,569 mtDNA data locations:

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>12345,HVR2,CR,HVR1
GATCACAGGTCTATCACCCTATTAACCACTCACGGGAGCTCTCCATGCATTTGGTATTTTCGTCTGGGGGGTG
TGCACGC
GATAGCATTGCGAGACGCTGGAGCCGGAGCACCCTATGTCGCAGTATCTGTCTTTGATTCCTGCCTCATTCTA
TTATTTA
... and so on ...
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WHAT CAN YOU DO WITH mtDNA?

Genealogists today want to know if DNA can solve a research problem. In some situations mtDNA provides conclusive evidence; in others it provides less strong evidence. The documented matrilineal lineage should be as deep as possible to make mtDNA most useful as genealogical evidence. The person to be tested must have a straight matrilineal descent—through women with no intervening men. A male can be tested, but the earlier ancestors must be all female back to the person of interest. See figure 1. Even without a specific problem to be solved, many are taking DNA tests to be part of this exciting technology and contribute to genealogical and scientific discoveries. More comprehensive tests (like the mtFullSequence) offer a better chance of finding a common ancestor in a genealogical time frame. If the mtDNA signature is rare the test provides stronger evidence for relationships within a more recent time frame. Even the low resolution mtDNA test provides strong evidence for some situations.

(1) Was Native American ancestry inherited down the direct matrilineal line? Native American ancestry can be indicated by specific haplogroups, but DNA cannot isolate to a specific tribe. The lower resolution mtDNAPlus test can often answer this question.

(2) Does a line descend from the first or second wife of a man? This requires that the two wives not be descended from a common matrilineal ancestor. If a man marries women sharing a matrilineal line the descendants will all be in the same mtDNA haplogroup. The lower resolution mtDNA test can sometimes answer this question, but the mtFullSequence test would be better.

(3) Which of several Johnson families in the area could be Mary Johnson's parents? You are a matrilineal line descendant of Mary Johnson who married John Richards. These ancestors are so far back that autosomal DNA may not be shared by cousins, but mtDNA will be shared by those in the matrilineal line. Research all potential Johnson families to locate living descendants who inherited mtDNA from the mother. If the descendants agree to test the mtDNA results can be compared to yours. If your mtDNA matches a test-taker from one line then focus on that Johnson family searching for more evidence indicating that couple are the parents of Mary. The mtFullSequence test would be best for this scenario.

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RESOURCES

This article is a short introduction to mitochondrial DNA. For information on tests offered by different companies see each vendor's web site and the International Society of Genetic Genealogists (ISOGG) Wiki pages "Mitochondrial DNA tests" and "mtDNA testing comparison chart."[1] For information on the reference samples, haplogroup nomenclature, and a graphic representation of the human mtDNA phylogenetic tree see *PhyloTree*.[2] For a compendium with links to most of the known information related to mtDNA and for information to analyze medical significance see *MitoMap*.[3] Remember to consult a medical or genetic professional as needed to understand the medical significance of mtDNA test results.

Debbie Parker Wayne, CG, CGL, is experienced using DNA analysis as well as more traditional techniques for genealogical research in Texas, the South and the West. She coordinates the genetic genealogy courses at several genealogy institutes and is the Texas State Genealogical Society's DNA Project director. She wrote the NGS *Continuing Genealogical Studies: Autosomal DNA* course and co-authored the first genetic genealogy workbook, *Genetic Genealogy in Practice*, published by NGS. See http://debbiewayne.com/ for more information.

[1]All URLs were accessed 24 January 2017.
"Mitochondrial DNA tests," Wiki, *ISOGG* (http://www.isogg.org/wiki/Mitochondrial_DNA_tests). "mtDNA testing comparison chart, " Wiki, *ISOGG* (http://www.isogg.org/wiki/MtDNA_testing_comparison_chart).
[2] Mannis van Oven, "PhyloTree.org," *PhyloTree* (http://www.phylotree.org/).
[3] "MitoMap: A Human Mitochondrial Genome Database," *MitoMap* (http://mitomap.org/).

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