International Journal of Sciences

Research Article Volume 5 – December 2016 (12)

The Mitochondrial DNA Mitotype of Sainte Marie-Madeleine

Gérard Lucotte[1]

[1]Institute of Molecular Anthropology, 75 005 Paris, France

Abstract : We have extracted *HVR1* (HyperVariable Region 1) mitochondrial DNA (mtDNA) sequences of Ste Marie-Madeleine (3?-63?) , from one capillary bulb of one of her hairs. These hairs are conserved in a reliquary that is exhibited in the St Maximin basilica. *HVR1* sequences show, reproducely twice, nine mutations : 16129G, 16187C, 16189T, 16223C, 16224C, 16230A, 16234T, 16278C and 16311T. The corresponding haplogroup is **K**, sub-clade **K1a1b1a**. As this sub-clade is the mtDNA genetic signature of ancient Jews, that confirms the Pharisian maternal origin of Marie-Madeleine indicated in some traditions.

Keywords: Mitochondrial DNA haplogroup ; Marie-Madeleine ; mutations in the hypervariable region 1.

INTRODUCTION

Ste Marie-Madeleine (3?-63?) is the most abundantly cited (at least twelve citations, without taking in account some repeats) women in the four Gospels.

According to the French "Tradition des Saints de Provence", she landed on (in 43?) the French (the Gaule at this era) Mediterranean shores, in a region corresponding to the current part of Les Saintes-Maries-de-la-Mer ; she (and her companions) attained further the towns of Marseilles and Aix, where they evangelised the French region of the Provence. She retired after, during thirty years, in a cave of la Sainte-Baume. She died (in 63?) and was buried in the currently named village of Saint-Maximin-la-Sainte Baume.

Some relics (bones and hairs) of Marie-Madeleine were conserved in the Saint-Maximin basilica, where a large lock of Marie-Madeleine's hairs is arranged in a dedicated reliquary. We have obtained some hairs (they are cut hairs, of red colour) of this lock, for scientific purposes (microscopic examination and chemical analysis). One of these hairs had a capillary bulb ; we have extracted DNA from this bulb, that permits us to obtain the corresponding mitochondrial DNA (mtDNA)

MATERIALS AND METHODS

The Hairs

Marie-Madeleine's hairs are arranged, at the interior of a brass-made reliquary (Figure 1), conserved in the Saint-Maximin basilica ; this photograph shows a voluminous lock of hairs (comprising several hundred hairs), linked together by two sorts (fine or thicker) of threads (probably silver-made). Several hair fragments were extracted from this lock by the basilica priest, and were loaded further on sterile scotch tapes (Figure 2). The first aim of this study consisted of the detection , by microscopy, of capillary bulbs from these hair fragments.

Microscopy and Elementary Analysis

Optic and electronic microscopy were realized by scanning electron microscopy (SEM), with the FEI model Quanta 250f FEG apparatus (Laboratoire d'Analyses physico-chimiques, UTC de Compiègne, France) ; both LFD (Large Field Detector) and CBS (Circular Back Scattering) procedures were used. This apparatus is equipped with a probe (Bruker model X-flash 6/30), that permits elementary analysis by EDX (Energy Dispersive X-rays).

DNA Extraction

Genomic DNA was extracted from the bulb using a standard method (0.5M EDTA, sarcosyl 20% and proteinase K 10 mg/ml), and purified using a commercial kit (Nucleospin^{+}kit ; Macherey-Nagel, Duren, Germany) in accordance with the manufacturer's instructions.

DNA extraction was performed independently in an isolated Laboratory (previously used mainly for work with human DNA), dedicated to working with ancient DNA (a-DNA).

Published at: http://www.ijsciences.com/pub/issue/2016-12/
DOI: 10.18483/ijSci.1167; Online ISSN: 2305-3925; Print ISSN: 2410-4477

Gérard Lucotte (Correspondence)
lucotte@hotmail.com
+06 98 82 92 61

Gender Detection
The amelogenin test was realized on the extracted genomic DNA, according to classical methods.

Amplification of the mtDNA Hypervariable regions
The mtDNA sequence intervals for *HVR1* and *HVR2* (Hypervariable regions 1 and 2) were amplified by PCR (Polymerase Chain Reaction) with primers F15971 and with primers F15971 and R16410 and with primers L15 and H484 , respectively. For each PCR, the DNA extract of the bulb specimen was amplified in a 15 µl reaction mixture : 2 mM $MgCl_2$, 50 mM KCl, 10mM Tris / HCl pH = 9, 0.1% Triton X-100 , 0.2mM each DNTPs, 0.1 µM each primer, and 2.5 U of DNA polymerase (Ampli Taq Gold ; Applied Biosystems, Foster City, USA). The amplifications were carried out with an initial denaturation at 95°C for 6 min., followed by 30-35 cycles at 95°C for 1 min., 55°C for 6 min., and 72°C for 1 min.

As for our previous works on ancient DNA (**1**), PCR procedures were performed in a sterile hood, in accordance with standards for a-DNA work, with regular decontamination measures and all precautions taken to avoid any risk of contamination by modern DNA molecules.

HVR1 Sequences
PCR products were purified from agarose gel (QIA-Quick PCR purification kit ; Qiagen CA, USA). Both strands of the amplified mtDNA fragments removed from agarose slides were directly sequenced (Big Dye Terminator Cycle sequencing kit ; Applied Biosystems) and separated (ABI PRISM 3130 Genetic Analyser ; Applied Biosystems).

Mutations detection
The sequences obtained were aligned on the Revised Cambridge Reference Sequence (**2**), to identify the presence of mutant sites. Seqscape software (Applied Biosystems) and Clustal Analysis (http://www.clustal.org) were used for pairwise alignments.

I was the unique experimenter in the present DNA procedure. My own mtDNA mutation in *HVR1* is 16298C only.

RESULTS
It is the 10^{th} studied hair only that shows a bulb at its basis ; the other finest extremity of the hair fragment number 10 shows the characteristic pattern of a wrenched-hair.

The SEM photograph of Figure 3 (and the corresponding optic photograph) depict the external anterior part of this bulb. For unknown reasons, the hair number 10 extremity at the bulb was natively cut. This hair extremity is enlarged, with an abrupt square end where we can see a pore (corresponding to the inferior opening of the medullar canal).

The global aspect of this bulb is that generally observed for all the other hairs studied : they are desiccated hairs, without well-observable scales and where the longitudinal crests of the keratin matrix of the hair cortex are clearly visible. But, at the surface, we can see at least five scraps corresponding to residual parts of the internal (sclerified) epithelial gain that covers the bulb.

Figure 4, in CBS, shows an enlarged (1 200 x) photograph of the bulb. Twenty particles of dense matter are visible at its surface. Table 1 characterizes , by EDX analysis, each of them : all (but particles numbers 3 and 14, that are dense hair fragments) are mineral / metallic particles. Such a preliminary analysis is important, because it permits us to verify that there is no evidence at the bulb surface of skin debris [**3**] - originating potentially from another individual-that could contaminate the sample.

Table 1. EDX analysis of particles shown on the bulb basis (shown on Figure 4).

Particles numbers	Particles compositions
1	calcite
2	iron oxide
3	hair fragment
4	calcite
5	copper oxide
6	clay+gold
7	clay+gold
8	calcium carbonate
9	calcium carbonate
10	clay+iron
11	clay
12	clay
13	clay

14	hair fragment
15	potassium phosphate
16	potassium phosphate
17	clay+gold
18	calcium carbonate
19	clay+iron oxide
20	clay+iron oxide

The photograph (1 200x) of Figure 5 shows the bulb upper part. It shows that this bulb is hollow, with a voluminous aperture corresponding to the upper part of the medullar canal.

Figure 6 shows an example of an EDX-analysis of the bulb. Carbon and oxygen peaks correspond to the organic matter, and the sulphur peak to keratin ; other peaks of calcium , potassium , silicium , aluminium, magnesium, phosphorus and sodium correspond to various mineral / organic deposits.

Results concerning mtDNA HVR1 sequence.
The natively cut bulb was isolated by micro-manipulation in sterile conditions. The approximate quantity of total genomic DNA obtained from the bulb is about 250 pg.

The amelogenin test practiced on this genomic DNA shows that it corresponds to the chromosomal XX formula of a woman.

Starting from the genomic DNA, we have realized (Table 2) three successive DNA essays (varying the hybridisation temperatures, the stringency of the PCR reactions and the number of cycles), in order to obtain *HVR1* and *HVR2* amplifications of the mtDNA sequences.

Table 2. DNA results obtained from the three essays (A : adenine ; C : cytosine ; G : guanine ; T : thymine).

Essays	**HVR1 amplification**	**HVR1 mutations**	**HVR2 amplification**
1rst essay	-		-
2nd essay	+	16129G, 16187C,16189T, 16223C, 16224C, 16230A, 16234T, 16278C, 16311T	-
3rd essay	+	16129G, 16187C, 16189T, 16223C, 16224C, 16230A, 16234T, 16278C, 16311T	-

We were unable to obtain any *HVR2* sequences . In the second and the third essays, we obtained *HVR1* amplification products (nucleotide positions from 16025 to 16359). A total number of nine mutations were detected, reproductively, in the two *HVR1* mtDNA sequences ; these nine mutations are : 16129G, 16187C, 16189T, 16223C, 16224C, 16230A, 16234T, 16278C and 16311T. That number of nine mutations , compared to the reference sequence, indicates an ancient human mtDNA haplogroup.

Presence of the 16224C (and 16311T) mutations determinates the mtDNA haplogroup **K** (http://www.phylotree.org), and that of the 16234T mutation indicates its **K1a1b1a** sub-clade. This panel of these nine mutations detected in *HVR1* is conform to those already obtained by Family Tree DNA (ftDNA) for some **K1a1b1a** living subjects (http://www.familytreedna.com/public/mtDNA K).

DISCUSSION
Working on authentic Marie-Madeleine's hairs, we have found a bulb for the 10th hair studied. Using the amelogenin test, we found that the DNA extracted from this bulb corresponds to that of a woman. Among the panel of the nine mutations we found in the mtDNA *HVR1* sequence, the 16224C and 16311T mutations (the basal node) determinate that this woman belongs to the **K** mtDNA haplogroup.

Overall, mtDNA haplogroup **K** is found in about 6% of the current populations of Europe and the Near East(http://www.oxfordancestors.com/content/view/35/55). But this percentage attains approximately 16% of the Druzes of Syria, and is elevated in Lebanon, Israel and Jordany ; in Palestinian Arabs, there is also a significantly elevated proportion (8%) of haplogroup **K**, that reaches at the level of 17% in Kurdistan (**4**).

Approximately 32-37% of people with Ashkenazi Jewish ancestry are of haplogroup **K**. This high percentage points to a genetic bottleneck occurring some 100 generations ago [**5**,**6**,**7**] . Behar *et al.* [**8**] report early finding of a significantly higher frequency of haplogroup K among Sephardic Levites (23%) and Sephardic Israelites (13%), probably the

highest frequency of **K** found among any (European) population. This may indicate that some of Ashkenazi haplogroup **K** is , in fact, of Israelite origin.

One of the greatest interest of the Behar *et al*. 2006 paper [**5**] on the maternal ancestry of Ashkenazi Jewry is that they traced back Israelite ancestry to only four women, carrying distinct mtDNA groups that are virtually sub-clade of the **N** haplogroup and three sub-clades of the **K** haplogroup : **K1ab1a**, **K1a9** and **K2a2a**.

K1ab1a sub-clade is marked by two – coding transitions (at the 10978 and 12954 sites) of the mtDNA , and includes fourteen of the 121 (=11.6%, the major **K** sub-clade found) of the 121 complete mtDNA sequences described. Seven of these were reported in [**5**] for the first time, and are from Ashkenazi subjects, whereas the other seven were reported elsewhere [**9**] as forming a specific cluster named "**K1a**". The ethnicities or religious affiliations of these last seven subjects are not available , but they were all collected in United States and shared the mtDNA control-region mutations with the Ashkenazi samples.

Newer researches have further updated the phylogenetic tree of haplogroup **K** sub-clades [**10**] . The **K1a1b1a1** sub-clade has yet to be approved and does not appear in the Build 17 Phylotree of February 18th, 2016 ; but it is well admitted that the characteristic mutations of the **K1a1b1a1** sub-clade are : 10978G, 12954C and (in *HVR1*) 16234T. As this last mutation is present (see table 2) in the *HVR1* mtDNA sequence of the women of the **K** haplogroup whose genomic DNA were extracted from her 10th-studied hair capillary bulb, we conclude that she is of the **K1a1b1a1** sub-clade. The current version 3 of van Oven's phylotree (http://www.phylotree.org) defines **K1a1b1a1** by the highly polymorphic 114 site in *HVR2*, sites 10978 and 12954 in the coding region, and the 16234 site in *HVR1*. This is supported by a growing number of Genebank (index.php?S=Genebank&item_type=topic) samples. However, the 12954 site is not needed to define **K1a1b1a** as of 2013 and, mentioned above [**10**] , is used now to define **K1a1b1a1**.

According to Jacques de Voragine [**11**], in his famous Middle-Ages story (but based on previous oldest traditions), Marie-Madeleine was of double royal lineages : her father was named Syrus and her mother Eucharie (or Euchérie) ; etymologically "Syrus" designates probably the Syrian people, and "Euchérie" (latinisation of the greek term Eucharie)means graceful.

Syrus was previously a Syrian Prince (or King). Other sources indicate that he was converted to Judaism (under the name of "Syrus the Yaïrite") and that he was the archipriest that officiated in the Capharnaüm synagogue. Marie-Madeleine's mother Eucharie descended from a rich Pharisian family, possibly of the Davidic lineage.

Estimates of the age of the **K1a1b1a** sub-clade vary, depending on the mutation rates adopted in the calculations and on historical population sizes. The age of **K1a1b1a** has been estimated at 4,800±3,600 years ago, according to the Genographic Project(index.php?s=Genographic%20%Project&item_type=topic). The **K1a1b1a** sub-clade is under the **U'K** haplogroup and descends from **K1a1b1,** which is thought to be an 11, 500-year-old European sub-clade of mostly non-Jewish origin.

A difficulty estimating Ashkenazim lineages is that they have undergone severe founder effects during their history [**12**], drastically altering the frequencies of genetic markers and distorting the relationship with their ancestral populations. There were Diaspora communities throughout Mediterranean Europe and Near-East for several centuries prior to the destruction of the Second Temple in Jerusalem in 70, and some scholars [**13**] suggest that their scale implies proselytism and wide-scale conversion.

There are two important questions with respect to the geographic origin of the Ashkenazi founding lineages [**5**] : **i**. Were these lineages a part of the mtDNA pool of a population ancestral to ashkenazi Jews in the Near-East, or were they established within the Ashkenazi Jews later in Europe as a result of introgression from European groups? **ii**. Were did the lineages expand? The observed global pattern of distribution renders very unlikely the possibility that the **K1ab1a** lineage (and the three aforementioned founder lineages) entered to the Ashkenazi mtDNA pool *via* gene flow from a European host population : in databases of *HVS1* sequences of British, German, French and Italian subjects, this Ashkenazi sample lineage sequence was not observed [**14**, **15**, **16**] . Furthermore, the non-Ashkenazi Jewish populations (Iberian Sephardims) sharing the Ashkenazi mtDNA haplogroup-lineages **K** turn out to be from Jewish communities that trace their origins to the expulsion from Spain in 1492 ; either a shared ancestral origin of the two groups may explain the sharing of these maternal lineages [**5**] , albeit at low frequencies, in North African and Near Eastern Jews supports a common Levantine ancestry. Moreover, the **K1a1b1a** –sister lineages (**K1a9** and **K2a2a**) which share with it a common ancestry at the internal nodal level of sub-clade **K1a1b1** , is found in Portugal, Italy, France, Morocco and Tunisia. All that reveals that this particular limb of the haplogroup **K** phylogenetic tree is of a wider Mediterranean presence and origin [**5**].

CONCLUSION

Because of its relatively little size compared to that of autosomal DNA, of the compaction of this DNA molecule that favorizes its conservation, and of its existence in multiple copies in each cell, mtDNA is since many times the favourite tool of the paleogeneticians working on fossils and ancient specimens. The most usable starting material is generally bones. A recent study [**17**] that had something to do with the present work on the Judean origins of Marie-Madeleine concerns sixty-three skeletons of the sites of Tell Halula, Tell Ramad and Dja ' de El Mughara (dated 8,700-6,600 B.C) that were analyzed. Fifteen validated mitochondrial DNA profiles were recovered ; comparisons with three ancient datasets allowed the authors to identify **K** (and **N**) – derived mtDNA haplogroups whose signature would have reached both the Iberian coast and the Central European plain.

The pioneer study in "biblic mtDNA research" is that published in 2001 by Vernesi *et al.* [**18**] on St Luc. According to historical sources, the Luke evangelist was born in Antioch, the Roman province of Syria, and died in Thèbes at 84 years old, around 150 A.D. His body was initially buried in Thèbes, but then transferred to Constantinople during the reign of Emperor Constantius, and after to Padua (Italy) at an unspecified time. The authors obtained the mtDNA by extraction from a canine tooth root originating from the body contained in the marble sarcophagus of Padua, body traditionally attributed to Luke. In the mtDNA-control region sequences, they found a cluster of mutations (mainly an **Alu** I cut at position 7025) that characterize the named "**pre-HV**" haplogroup (that is relatively common in current populations of the Mediterranean region). By comparison with modern typed samples from Syria and Greece (and with those samples from Anatolia that were already available in the literature), the authors have shown that the Luke mtDNA sequence have "a nearly 3-fold higher probability to come from a Syrian than from a Greek individual."

In the present study we report the mtDNA haplogroup of Marie-Madeleine , obtained extracting mtDNA from one bulb of one of her hair. The 16224C mutation found in the *HVR1* mtDNA sequence established that she belongs to the **K** mtDNA haplogroup ; as we found also the 16234T mutation in its, she belongs to the **K1a1b1a** sub-clade of **K** (as named as **K1a1b1a1** in the current nomenclature). That constitutes the Marie-Madeleine mitotype. As demonstrated in [**5**], the **K1a1b1a** sub-clade is a genetic signature of Jewry ; so, the result we obtain concerning the **K1a1b1a** sub-clade of Marie-Madeleine sustains her Jewish ancestry originating from her mother (as described in some traditions).

The **K1a1b1a** lineages (within which the **K1a1b1a** sequences nest) are solely European [**10**], that indicating an ancient European ancestry. Otherwise that Ashkenazics, the **K1a1b1a** sub-clade of ancient Jews, is also present (at low frequencies) in Spanish-exile Sephardic Jews [**6**] ; it is not seen in Libyan Jews, who are known to have a distinct Near Eastern ancestry. To our knowledge Djerban Jews [**19**] , with a similar history of that the Libyans, have not been tested for mtDNA.

Researches on membership of the Davidic lineage, a strict Y-chromosome concept [**20**] from the genetic point of view , cannot be applied (even indirectly) to a woman.

List of abbreviations.

a-DNA : ancient DNA ; mtDNA : mitochondrial DNA ; *HVR1* : HyperVariable Region 1 of the mtDNA ; **K** : the mtDNA haplogroup found ; **K1a1b1a** (and **K1a1b1a1**) : the sub-clade of **K** found. PCR : Polymerase Chain Reaction ; SEM-EDX : Scanning Electronic Microscopy – Energy Dispersive X-rays ; LFD : Large Field Detector ; CBS : Circular Back Scattering.

Acknowledgments.

I thank Florian Racine, the priest of the St-Maximin-la-Sainte-Baume, who furnished to me Marie-Madeleine's hairs contained in the reliquary. I acknowledge also A. Salmon, C. Riondel and B. Pey, who facilitate such a translation. F. Racine is also acknowledged for his initial financial support. This study is included in our program on scientific investigations developed at the Institute of Molecular Anthropology concerning the vestimentary Christ relics (Argenteuil Tunic, Turin Shroud, Prüm Sandal and so on).

Additional information : The *HVR1* mtDNA sequences of the Marie-Madeleine hairs are also available on demand at : lucotte@hotmail.com

References

1. Lucotte G. A rare variant of the mtDNA HVS1 sequence in the hairs of Napoléon's family. *Investigative Genetics*, 2010, 1 : 1-6.
2. Andrews RM, Kubacha I, Chinnery PF *et al*. Reanalysis and revision of the Cambridge reference sequence for human mitochondrial DNA. *Nature Genetics*, 1999, 23 : 147 (doi : 10.1038/ 13779).
3. Lucotte G. Skin debris on the Face of the Turin Shroud : a SEM-EDX analysis. *Archaelogical Discovery*, 2016, 4 : 103-117.
4. Simoni L, Calafell F, Pettener D, Bertranpetit J, Barbujani G. Geographic patterns of mtDNA diversity in Europe. *American Journal of Human Genetics*, 2000, 66 : 262-278.
5. Behar DM, Metspalu E, Kivisild T *et al*. The matrilineal ancestry of Ashkenazi Jewry : portrait of a recent founder event. *American Journal of Human Genetics*, 2006, 78 : 487-497.
6. Behar DM, Metspalu E, Kivisild T *et al*. Counting of the

founders : the matrilineal genetic ancestry of the Jewish diaspora. PLOSone, 2008 (doi.org/ 10.1371/ journalpone.0002062).

7. Feder J, Ovadia O, Glaser B, Mishmar D. Ashkenazi Jewish mtDNA haplogroup distribution varies among distinct subpopulations : lessons of population substructure in a closed group. *European Journal of Human Genetics*, 2007, 15 ; 498-500.
8. Behar DM, Thomas MG, Skorecki K et al. Multiple origins of Ashkenazi Levites : Y chromosome evidence for both Near Eastern and European ancestries. *American Journal of Human Genetics*, 2003, 73 : 768-779.
9. Herrnstadt C, Eloon JL, Fahy E *et al.* Reduced-median network analysis of complete mitochondrial DNA coding-region sequences for the major African, Asian, and European haplogroups. *American Journal of Human Genetics*, 2002, 70 : 1152-1171.
10. Costa MD, Pereira JB, Pala M *et al*. A substantial prehistoric European ancestry amongst Askenazi maternal lineages. *Nature Communications*, 2013, 4 : 2543 (doi : 10.138/ncomms3543).
11. Voragine. La légende dorée. Gallimard Ed., 2004 (chap. 92).
12. Ostrer H. Legacy : a genetic history of the Jewish People. Oxford University Press Ed., 2012.
13. Irshai O. *In* The illustrated history of the Jewish people. De Lange N. Aurum Press Ed., 1997.
14. Baasner A, Schafer C, Junge A, Madea B. Polymorphic sites in human mitochondrial DNA control region sequences : population data and maternal inheritance. *Forensic Sciences International* , 1998, 98 : 169-178.
15. Lutz S, Weisser HJ, Heizmann J, Pollak S. Location and frequency of polymorphic positions in the mtDNA control region of individuals from Germany. *International Journal of Legal Medicine*, 1998, 111: 67-77.
16. Pfeiffer H, Foster P, Ortmann C, Brinkmann B. The results of an mtDNA study in 1200 inhabitants of a German village in comparison to other Caucasian databases and its relevance for forensic casework. *International Journal of Legal Medicine*, 2001, 114 : 169-172.
17. Fernandez E, Perez-Perez A, Gamba C *et al*. Ancient DNA analysis of 8000 B.C. Near Eastern farmers supports an early Neolithic pioneer maritime colonization of Mainland Europe through Cyprus and the Agean islands. *PLOSone*, 2014 (dx.doi.org/10.1371/journal.pgen.1004401).
18. Vernesi C, Benedetto G, Caramelli D *et al.* Genetic characterization of the body attributed to the evangelist Luke. *Proceedings of the National Academy of Sciences*, 2001, 98 : 13460-13463.
19. Lucotte G, David F, Berriche S. Haplotype VIII of the Y chromosome is the ancestral haplotype in Jews. *Human Biology* , 1996, 68 : 467-471.
20. Thomas MG, Skorecki K, Ben-Ami H *et al.* Origins of Old Testament priests. *Nature*, 1998, 394 : 138-140.

Figure 1. The interior of the reliquary. H : hairs ; T : background textile ; P : the paper ; W : red sealing wax ; Ⓑ: the bone. On the inscription : …Capill. S. 1Oss.S.M.M. (hairs on one bone of Sainte Marie-Madeleine).

Figure 2. Hairs numbers 4 to 10 on the scoth tape. The ⓐextremity of hair number 10 corresponds to a hair bulb.

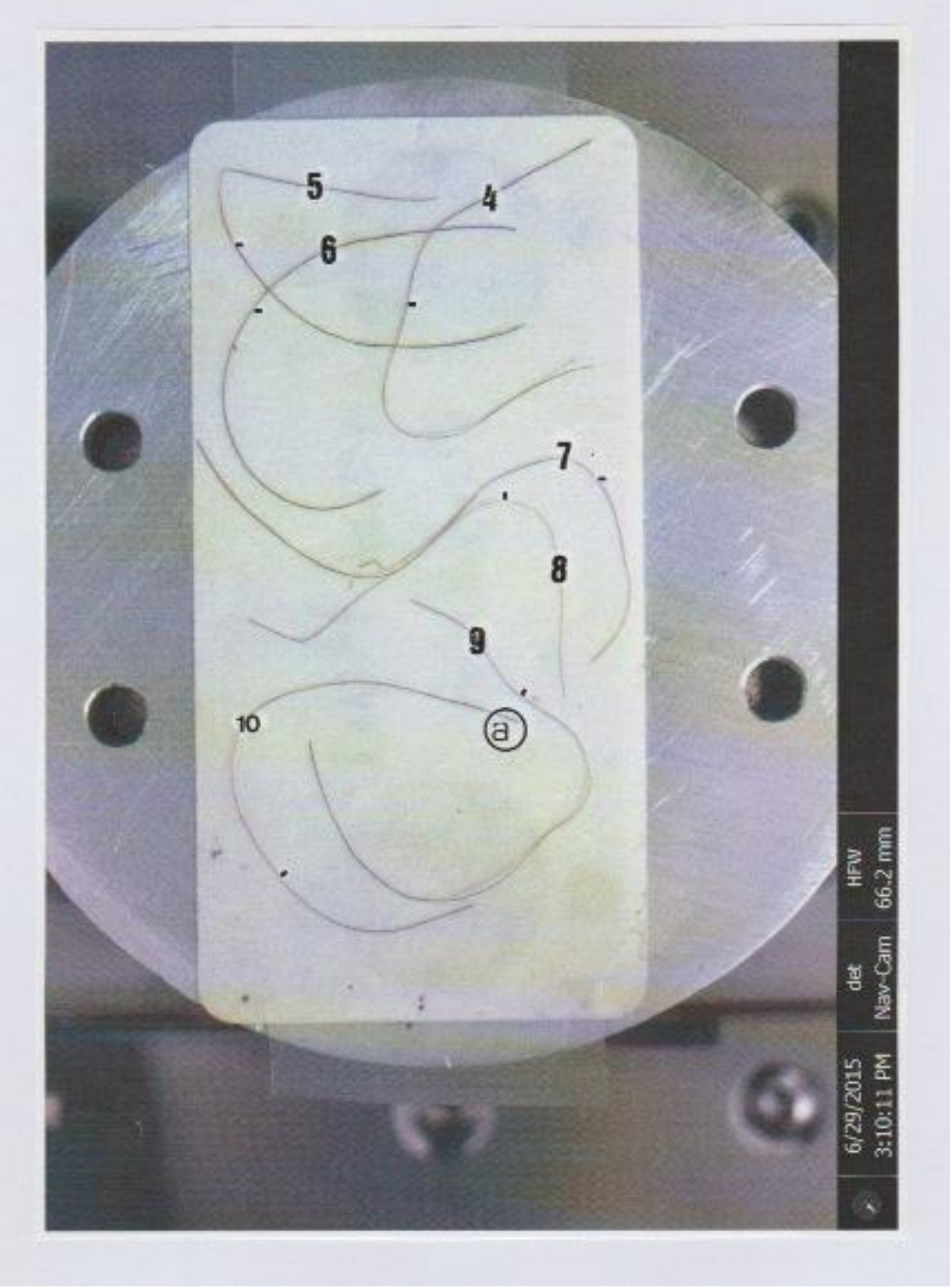

Figure 3. Above : a SEM photograph (300x), in LFD, of the hair number 10 basis. C ⇒C indicates the natively cut section of the hair bulb (B) ; P : scraps 1, 2, 3, 4 and 5 : scraps of organic matter. Below : optic photograph (250x) of the hair (H) basis and the bulb ; the hair and the bulb are of the same (red) colour.

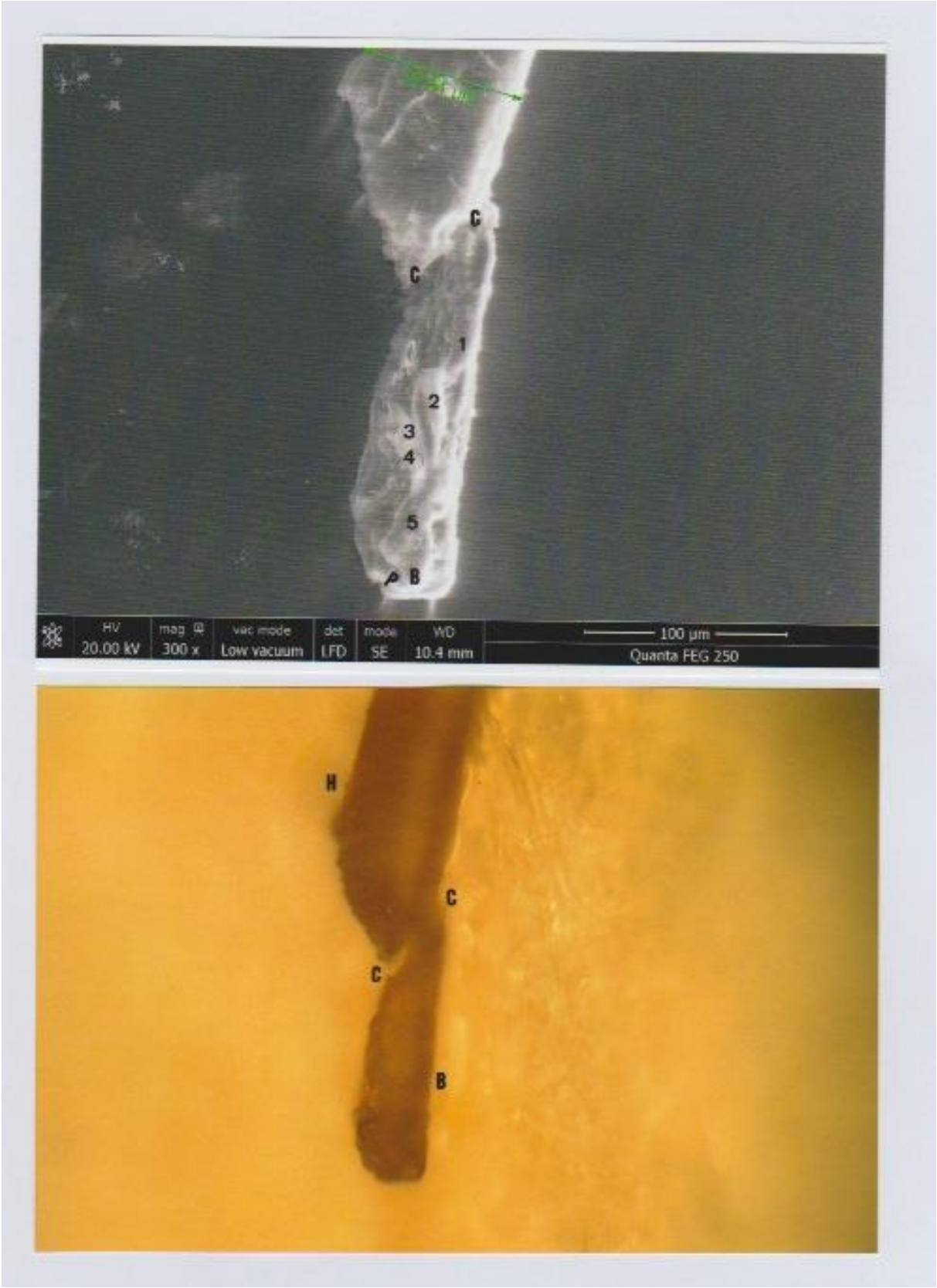

Figure 4. SEM photograph (1 200x) , in CBS, of the bulb basis. Numbers 1 to 20 indicate particles analysed by EDX.

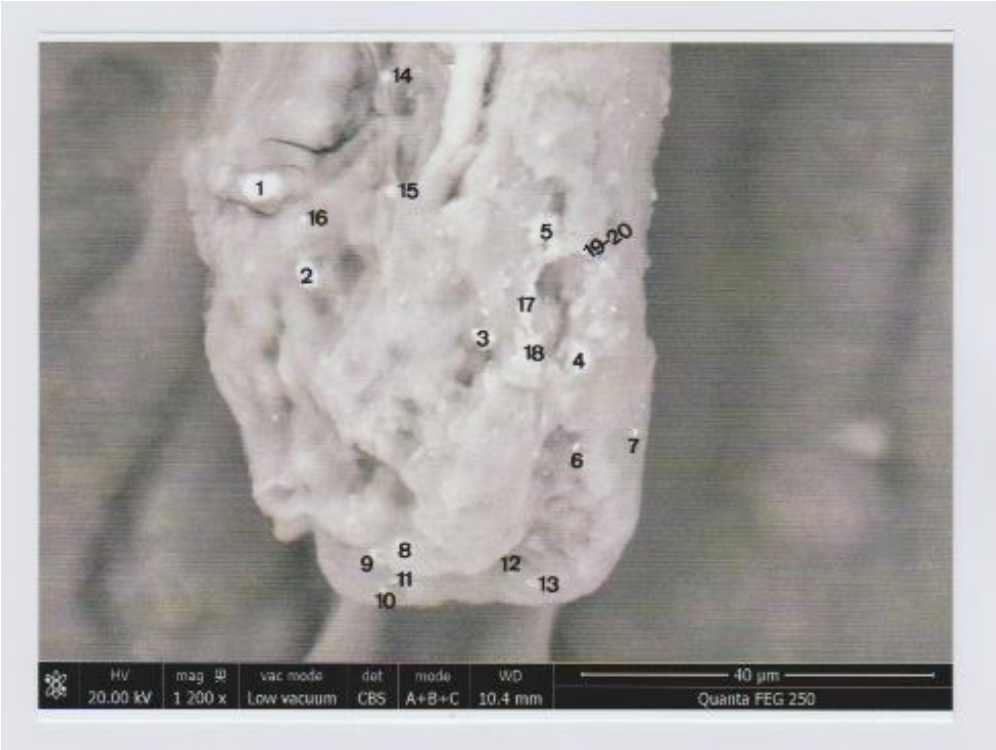

Figure 5. SEM photograph (1 200x), in LFD, of the bulb upper part, under the natively section. C : crests of the dessicated hair cortex in the hole (H). G1 : the scrap number 1 ; G2 : upper part of the scrap number 2 ; G3-5 : other little scrap debris.

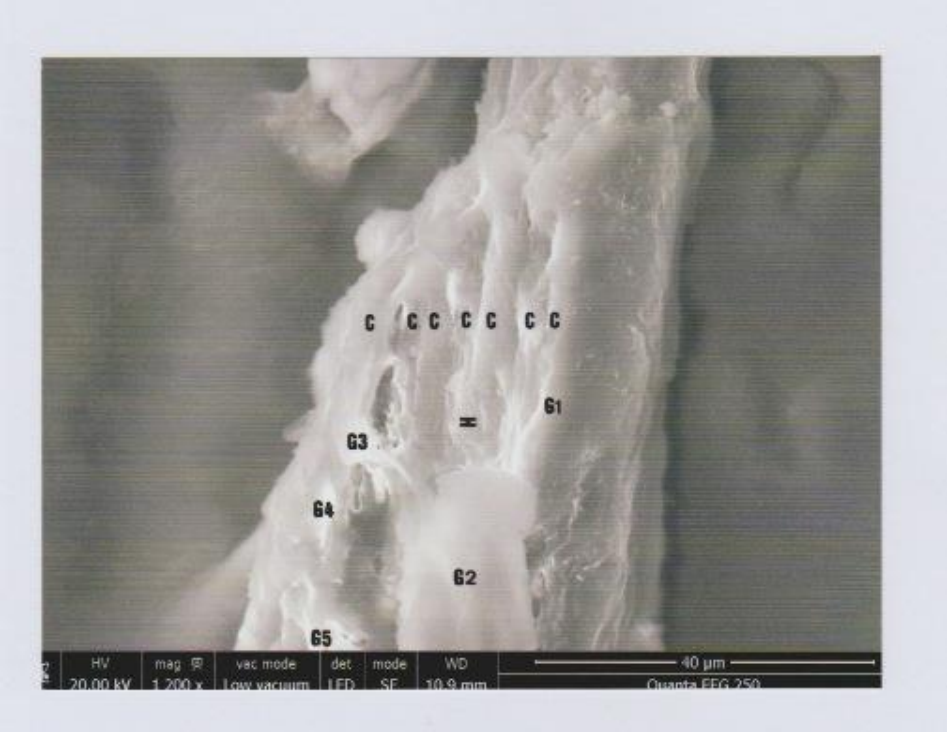

Figure 6. Above : SEM photograph (1 200x), in LFD, of the bulb basis lower part ; H : hole of the lowest medullar canal aperture. Below : EDX analysis , at the black point indicated. C : carbon ; O : oxygen ; Na : sodium ; Al : aluminium ; Si : silicium ; P : phosphorus ; S (two peaks) : sulphur ; Ca : calcium.