The Shroud Blood Science of Dr. Pierluigi Baima Bollone: Another look at potassium, among other things

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Although probably best known for his blood typing studies on the Shroud, it is worth noting that Dr. Pierluigi Baima Bollone conducted a series of chemical and immunological studies in the characterization of Shroud bloodstains, similar to those performed by Heller and Adler. [Translated from the book jacket shown above:] "Dr. Pierluigi Baima Bollone, 76, a surgeon, for over 40 years, teacher of Forensic Medicine in the Faculty of Medicine and Law at the University of Turin-and now professor emeritus, continues to contract in his courses. He is the author of a successful Handbook of Forensic Medicine adopted in various Universities, now in its fifth edition, and 161 scientific publications. He has also written 24 books."

Below, several specifics regarding Baima Bollone's findings relative to those of Heller and Adler are discussed. Much of the quoted material is taken from his presentation entitled "The Forensic Characteristics of the Blood Marks" from The Turin Shroud-past, present, and future, an International Scientific Symposium held in Turin March 2-5, 2000. Direct quotes from Dr. Baima Bollone are **bolded**.

The first endeavor to scientifically evaluate the nature of the bloodstains on the Shroud began in 1973 by members of the "commission of experts", which included G. Frache, E.M. Rizaati, and M. Mari. Their results were negative, although the scientists would conclude that "the negative answer to the investigations conducted does not permit an absolute judgment of the hematic nature of the material under examination." In a 1981 paper by Baima Bollone, entitled "Indagini Identificative Su Fili Della Sindone", he describes his own initial studies testing for the presence of hemochromagen, which were also negative, corresponding to the work of Frache, et al. In subsequent studies he would use different methods; discussing this at the 2000 conference, he states, "**The initiative to conduct new tests developed a few years later, during the laboratory examination of threads extracted from the Shroud and adhesive tape samples. Under the fluorescence microscope and using the Dotzauer and Keding method on the same samples I demonstrated the presence of heme/porphyrins. On the same material I obtained Teichmann crystals or hematine chlorohydrate with the usual procedures.**" In follow up studies, Baima Bollone would extend his chemical results using a series of immunological experiments, which tested positive for the presence of blood component markers. Immunological studies by Heller and Adler (albumin, immunoglobulin), and Garza-Valdes (hemoglobin, typing) would corroborate Bollone's findings.

Regarding the methods of sampling used in Shroud blood studies, Baima Bollone would comment in 2000, "I would like to point out immediately that the traces suspected of being "blood" are made up of effective deposits of material. This has meant that it has been possible to remove some of the traces using adhesive tapes or to study them directly on threads where they are deposited." This is a distinction between the two sets of studies: Heller and Adler primarily evaluated tape-lift samples, whereas Baima Bollone physically removed certain threads using forceps. He would further discuss that, "I have been astonished that in their search for traces of blood on the Shroud the STURP team preferred to use physics investigations, or at most surface sample-taking using adhesive tapes, rather than requesting to takes the traces in their materiality."

"After preliminary studies on bandages taken from an Egyptian mummy in order to optimize methods, in 1981 I centered research on the threads of the weft and warp taken in correspondence with the C9d area of the reference map (the so-called "belt of blood"), B12c (the sole of the left foot) and...(the sole of the right foot) of the feet of the Shroud. After optical and scanning electron microscope investigations, I managed by means of the energy extinction microspectrometer to ascertain the presence of Mg, Al, Si, S, Cl, K, Ca and Fe."

Here there appears to be a difference between the findings of Baima Bollone relative to those reported by the STURP scientists. In the article, "The origin and nature of blood on the Turin Shroud", Adler states that the blood is "very low in potassium", referencing the x-ray fluorescence studies of Morris and colleagues ("X-ray Fluorescence Investigation of the Shroud of Turin"). These specific tests were done at various places on the Shroud, including bloodstains, to help define if the elemental signature was more like paint or pigment? Or blood? Or other?

In the Morris studies the Shroud was sampled while mounted on a specially constructed frame. Bloodstains on the dorsal-foot and the side wound were analyzed. A spectrum of the side wound is presented in the Morris paper (which is also reproduced in Walter McCrone's book, together with a standard reference blood spectrum). Morris et al. state that, "Although no potassium was observed in any of the Shroud data, poor signal-to-noise ratios may preclude definite conclusions on this point" (see below).

In contrast to these findings, Baima Bollone and coworkers report in the evaluation of threads taken from the Shroud that the elemental profile, including K (potassium), is similar to that of normal blood. This work is described in the articles "La Dimonstrazione Della Presenza Di Tracce Di Sangue Umano Sulla Sindone" and "Indagini Identificative Su Fili Della Sindone". The "important blood peaks" labeled in McCrone's elemental analysis of a real blood profile (S, Cl, K, and Fe) are present in Baima Bollone's analysis. This is a very important point.

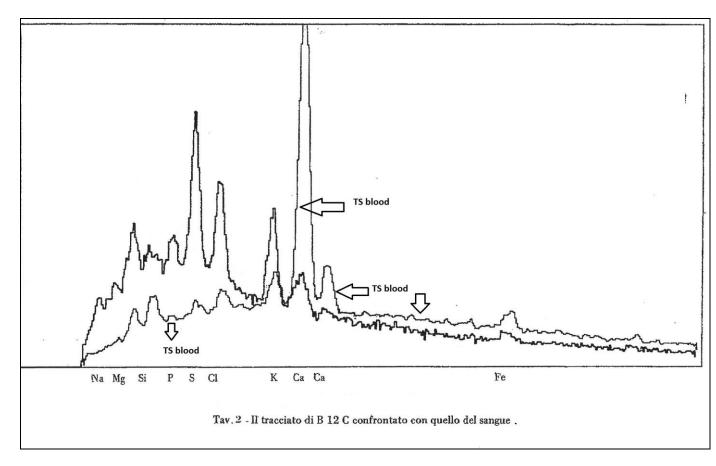
In the studies by Morris et al., X-Ray fluorescence (XRF) was used, while Bollone used Electron Dispersive Spectroscopy (EDS). The difference between EDS and XRF is the type of radiation hitting the sample. EDS uses an electron beam and XRF uses an x-ray beam, thus the results cannot be directly compared as such. However, both methods give the elemental composition of a sample and have their own limits, but, in fact, they are complementary.

Morris and coworkers were only allowed to perform their work on the Shroud itself; it is noteworthy that they could obtain valuable results. Among the normal major chemical species found in blood (see below), Aluminum, Silicon, Phosphorus and Sulfur could not be detected because their atomic number is lower or equal to 16. However Chlorine, Potassium, Calcium and Iron should, in theory, have been detected in blood areas. Given the very high amount of calcium thorough the entire Shroud, a small excess of calcium in blood areas could probably not be detected. They found an excess of iron in blood areas, consistent with the amount of iron in blood. There is no mention of Chlorine in Morris'paper.

Morris et al.were unable to detect the characteristic peak of potassium in their spectra of bloodstains. They added: "In these measurements [laboratory experiment with whole blood on a Whatman paper using the same XRF system than in Turin], we also observed potassium in addition to iron. The K [potassium] K alpha peak intensity was typically at least an order of magnitude smaller than the Fe K alpha." Given that and the very low signal to noise ratio of the TS spectra, it would be very unwise to conclude (like McCrone): "no potassium, no blood".

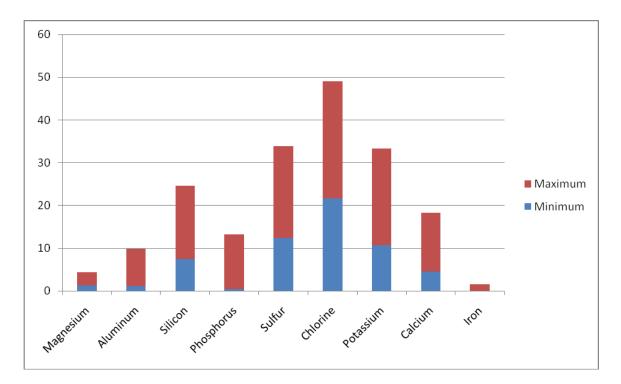
Baima Bollone used EDS on TS blood threads.

As an example, he got the following EDS spectrum:



From Dr Baima Bollone: comparison of EDS spectrum of TS blood (B12 C: sole of the left foot) and EDS spectrum of actual blood. There is a good match between actual whole blood and TS blood and potassium is there (arrows added to the original figure).

Dixon et al. studied the elemental composition of dried blood on cloth (Dixon et al., "A Scanning Electron Microscope Study of Dried Blood, 1976) using SEM-EDX. In this paper the elemental composition (the species and their relative amount) are given.



Mimimum and maximum values of elemental species (in %) in dried blood on cloth. From the SEM-EDX data in Dixon et al.

SEM-EDX gives the relative elemental composition. When one compares these results with those obtained by Baima Bollone, there is a very good match with "actual blood". This is also true for the TS blood, except for calcium and iron. Bollone's "TS blood" does contain much more calcium and iron than expected for actual blood. Since SEM-EDX analyses a volume of several micrometers, it is possible that the calcium (and iron) excess found in the "TS blood" spectrum is due to the high amount of calcium and iron bounded to the underlying fibers as shown by Heller and Adler.

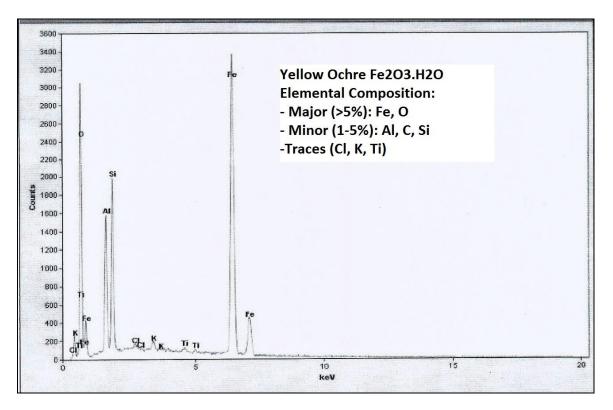
In any case (except for calcium), there is no doubt that the EDS spectra of blood material coming from blood areas of the Turin Shroud show the same elemental composition than that of actual blood. Not only are all of the expected elements present, but also their relative amounts are consistent with that of blood. In addition, no peak corresponding to species not pertaining to blood was found (for example Hg peak of cinnabar).

It is also important to note that Heller and Adler studied "globs" (blood aggregates) and fibrils from blood image using an EDS spectrometer ("A Chemical Investigation of the Shroud of Turin", 1981). They wrote: "The fibrils all show strong Ca and Fe signals. The globs all show Na, Mg, Al, Si, P, S, Cl, K [potassium], Ca and Fe ... Similar results were obtained by

J. Jackson and W.Ercoline in their SEM studies". Although no spectrum is shown, it is important to note that all of the elemental species of real blood were also found by Heller and Adler on "globs" (and perhaps red fibrils, although this is unclear), including potassium.

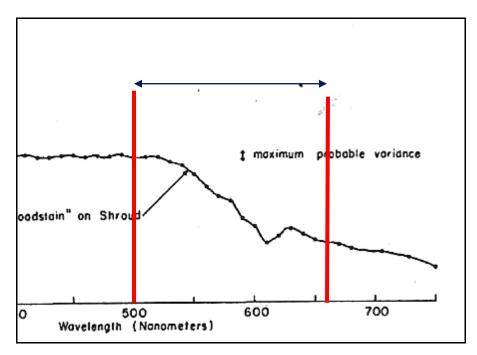
To summarize, the assumption that no potassium has been found in the blood stains is simply false. This assumption was only based on the Morris et al. paper and we have seen the strong limits of their in situ observations. On the other side, Bollone, Heller and Adler using another method found potassium in red material and blood threads. Bollone has provided evidence that the elemental composition of the TS blood effectively compares to that of actual blood.

McCrone thought that the blood was made of a mixture of red/yellow ochre with cinnabar in a collagen medium.



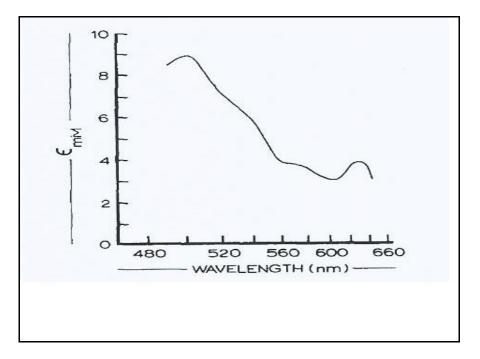
Typical SEM-EDX spectrum of pure yellow ochre (from McCrone particle atlas).

Iron, silicon and aluminum are the major species. Chlorine (a major element of blood) is only found as trace element. Even if one adds vermilion (artificial Hgs) or natural cinnabar (HgS and contaminants like quartz and calcite), it would be extremely surprising that the spectrum of such a mixture could match that of actual blood as does the TS blood spectrum. Another important issue relates to the reflectance spectra of blood. In "Physics and chemistry of the Shroud of Turin- A Summary of the 1978 Investigation" (Analytica Chimica Acta, 135 (1982) 3-49), Rogers et al. commented that "Heller and Adler have noted that there is no specific spectrum for blood per se: the spectral characteristics depend on the chemical state of the hemoglobin and also on its state of aggregation. They pointed out the strong resemblance of the Gilberts' "blood" data to those for perturbed acid methemoglobin, which is a chemical state of blood in which the iron in the hemoglobin has been oxidized. Cameron and George have published absorption spectra for acid methemoglobin in the range 480- 640 nm. These data strongly resemble the Gilberts' curves and even include the small absorption structure at about 630 nm".



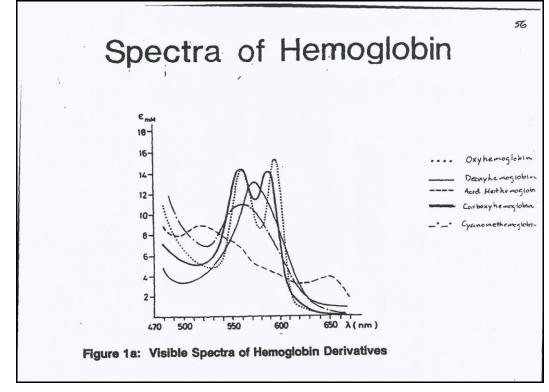
Apparent relative spectral absorbance of the mean of four bloodstained areas on the Shroud (Gilbert and Gilbert)

Here is the spectrum from Cameron et al., cited by Rogers:



Spectrum of human acidic ferrihemoglobin (Cameron et al.)

The claim that "there is no specific spectrum for blood per se" can be seen in the following figure:



In "Spectroscopy of burn wounds" by Afromowitz M.A (Ph D) and Callis J.D (Ph D), University of Washington, 1992.

Among the different spectra of hemoglobin, the spectrum of acid methemoglobin is unique. The spectrum of the bloodstains on the Shroud is consistent with that of acid methemoglobin found in the literature.

Bollone stated in 2000. "The forensic identification of the blood was obtained in 1981 by J.H. Heller and A. Adler. In 1980 they had already ascertained the presence of porphyrin, a pigment that enters among other things in the haemoglobin synthesis, in their samples." He continues, "The presence of human blood was subsequently confirmed by Canale in 1995 before conducting DNA research on some by threads I gave him, and by Leoncio Garza-Valdes, both on material of asserted but not proved origin from the Shroud, and on fragments of Shroud tapes from 1978 obtained from Adler."

No singular type of test in the evaluation of bloodstains is above error. Each test can result in a false positive. Each test can result in a false negative. It is the sum of the collective evidence of chemical and immunological data that convinced Dr. Baima Bollone (and Heller and Adler) that the "bloodstains" were composed of real blood. Although modern tests are typically more sensitive than many previous methods, the basic one-two approach for the detection of blood is still in use today: chemical testing for the identification of heme/hemoglobin, followed by immunological testing to identify the species from which the blood originates, and if desirable, the blood group and subgroup. (Reviewed in "Analysis of body fluids for forensic purposes: From laboratory testing to non-destructive rapid confirmatory identification at a crime scene", by Virkler and Lednev, 2009; and "Review: Biological evidence collection and forensic blood identification", by Castro et al., 2011).

Baima Bollone contributes a unique perspective in the study of the bloodstains of the Shroud, being trained in forensic science, and having evaluated the cloth at a very close range. He also discusses at some length the separation of cellular and fluid components in bloodstains relative to deposition, time of death, and clotting in his studies. As with all of the above-mentioned results, the interested reader is encouraged to consult the original sources of his articles and books for further information.

Baima Bollone would summarize in 2000 that, "In effect everyday haematological diagnostic investigations have allowed us to ascertain the incontrovertible presence of human blood, with all its characteristics, on the Shroud. All this proves and confirms that on the Shroud there are effectively real and complete bloodstains, conserved in their various components"