Christina Gram on the Gram Stain in Letters to Carl Julius Salomonsen, 1883–1884

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It has been a matter for discussion to what extent Christian Gram realized the significance of the staining method that he discovered in Friedländer's laboratory in 1883. Probably the most thorough analysis is that of Austrian (1) 20 years ago. Austrian reached the conclusion that neither Gram nor Friedländer gave any sign when Gram's paper appeared of having attached any importance to the observation that the tinctorial properties of the organism cultured by Friedländer were unique among those of the 20 cases of fatal pneumonia studied by Gram by his staining method. One cannot but agree with this conclusion based on the published literature.

However, recent studies in the Royal Library in Copenhagen have revealed nine letters (8) in Danish from Christian Gram written in 1883 and 1884 which demonstrate that Gram's understanding of the significance of his method went further than appears from his published papers, and the letters also suggest why he refrained from being more outspoken in his publications. Parts of these letters undoubtedly deserve to be known by all bacteriologists interested in the history of their science, and extracts are presented here in an English translation.

The letters were discovered during a systematic perusal of the correspondence of the Danish pioneer bacteriologist, Carl Julius Salomonsen (1847-1924).

In 1883 Salomonsen was appointed temporary lecturer in bacteriology at the University of Copenhagen, with the specific task to run courses in practical bacteriological techniques for medical students and others. Among the pupils attending Salomonsen's first course was Christian Gram, who had graduated 5 years previously and was then 30 years old. Gram had planned to study abroad when the course was finished, and Salomonsen gave him an introduction to Friedländer whom he knew from a visit to his laboratory in Berlin the previous year. Gram arrived in Friedländer's laboratory on 22 October 1883 and continued to work there until 20 March 1884. During this period he wrote six
letters to Salomonsen to report on his work and to tell about his life in Berlin. After his stay in Berlin Gram spent 5 or 6 weeks on holiday in Northern Italy and Switzerland, where he met Friedländer again in Lugano, and then went to Strassburg to work on ptomaines and digitoxin in Oswald Schmiedeberg laboratory. There he stayed until the beginning of August, when he returned to Copenhagen to attend the 8th International Medical Congress from August 10 to 16. From Strassburg he wrote another three letters to Salomonsen, partly to make arrangements concerning a paper he was going to read at the Congress (Salomonsen was one of the secretaries of the Congress).

All of the above-mentioned nine letters contain references to the Gram staining method or to Friedländer’s work on the pneumonia cocci. All parts of the nine letters which deal with these two subjects have been translated here from the original Danish into English. The remaining parts of the long letters that are mainly concerned with private matters and local medical gossip are not included in this translation, although they are interesting enough as a contribution both to an understanding of Gram’s personality and to medical and bacteriological history in general.

Gram’s language is youthfully facetious, and instead of speaking plainly he often uses exaggerated and awkward terms. I have tried as far as possible to retain his stylistic peculiarities in the translation.

Letter 1, dated Berlin, 5 November 1883

“. . . First and foremost, however, I want to thank you very much for your recommendation to Dr. Friedländer; he received me most cordially and has kindly taken care of me. Already on October 22 I settled with him and have since worked there every day. I help him to some degree with his work on the pneumonia cocci. Otherwise things are as they are with you: cultures on gelatin and potatoes and putting to death mice, guinea pigs and dogs. The pneumonia bug has now successfully been carried through many generations in culture and has produced pneumonia in the experimental animals, and next Friday Dr. Friedländer will read a paper on it in the Physiological Society. As I do not know how much Dr. F. wants to have leaked out to the public, I am not going to report any details. My work has mainly been on the microscopical demonstration of the bugs in the organs (in sections) and I have had the good luck to find what seems to be a very good method to stain the cocci while the tissue and the cell nuclei remain unstained. Dr. F. is delighted with the method; I take the matter more calmly and try to develop it further . . .

“. . . If you want some cover slips with pneumonia cocci or some cultures I can send you as many as you want . . .”

Letter 2, dated Berlin, 4 December 1883

“I beg to include for you partly some cover slip preparations of pneumonia cocci with capsules, partly some slides of my sections. Unfortunately, the slides from animals (cocci with capsules) are bad at the moment, since all the best ones have been given away, but the one included can be used. In contrast, my slides from humans are fine.

“I hope that you received a gelatin culture safely.”

Letter 3, dated Berlin, 18 December 1883

“. . . I have brought as much pressure to bear on Friedländer as possible . . . He has almost promised to talk about the pneumonia cocci and, anyway, we have agreed that if he does not come, I shall represent him and demonstrate the bugs. However, we will have to talk this matter over more closely since it will require some arrangements with cultures, etc. However, there is a difficult point in the story about these pneumonia cocci with capsules. After pathological-anatomical examinations of fresh and old (spiritus) specimens from pneumonia cases I have become sceptical, but as yet I dare not commit myself further.

“My activity down here is almost unchanged, I cultivate various bugs in tubes and examine all kinds of specimens by the aid of my “excellent” iodine method . . . .”

Letter 4, dated Berlin, 2 January 1884

“. . . Next, it can be reported that just now Friedländer is not going to the congress. Honestly, I do not believe either that he is coming since he will be on leave of absence in March and April.

“I have finished my studies on the famous iodine decolorization method which, I assume, will soon be published; however, in this connection there is a difficult point which Friedländer does not quite like. I will have to tamper a little with his work, especially his “capsules.” It will be somewhat difficult to do that gently. It seems as if there are at least two varieties of pneumonia cocci that look very different . . . .”
Letter 5, dated Berlin, 22 January 1884

"... Apart from this, I have not much new to report. Only the sad message, which I have previously suggested, that the capsules of the pneumonia cocci seem to be doubtful; at least, so far I have only seen them in the experimental animals injected from one particular case in which cocci with capsules occurred. In 21 cases of pneumonia cocci without capsules occurred. On the whole, we seem to get curious results with the pneumonia. Friedländer is not pleased and has asked me temporarily to withhold a paper on the iodine method, as it would have to contain certain disclosures. It also appeared to me that his paper was not quite good; he only had cultures from one single case."

Letter 6, dated Berlin, 28 February 1884

"... Only that much I want to tell you: my Berlin paper will appear in the next issue of "Fortschrifite" under the euphonious title "Über die isolierte Färbung der Schizomyzetten in Schnitt- und Trockenpräparaten." The fact is that I have developed and extended my "famous" iodine method to be valid for almost all Schizomyzetes, so that one obtains equally beautiful (and durable?) double-colored slides as in the well-known tubercle bacillus slides.

I have by now examined a whole series of microorganisms and yesterday I demonstrated my method and my results in the Reichsgesundheitsamt, where the natives became "en-chanted" and immediately started painting their bugs with aniline-gentiana-violet and iodine. By the way, Rasmussen has been initiated in the method and carries some slides with him to Denmark; he can give you information on the "epoch-making" discovery if you are interested."

Letter 7, dated Strassburg, 3 June 1884

"... Your critical remarks have not hurt my pride; admittedly I had overlooked that Weigert's (not Koch's) method had been published. I had seen several slides in the Reichsgesundheitsamt, but there they (Löffler) did not know the method of Weigert, which, however, had only been applied to anthrax bacilli; also, as far as I have learned through Weigert himself, he has not used it for other specimens than those mentioned. Anyway, Weigert's method must have been fairly completely forgotten, since Löffler with mortification remarked that they had "wunderschöne" slides of anthrax kidneys from Weigert, but they did not know the method.

"You were kind enough to invite me to deliver some twaddle at the congress—and I am willing enough, I suppose—only at that time I had absolutely nothing to report. I only had offered—through Friedenreich—to demonstrate some slides with "bacteria" (more correctly Schizomyzetes) stained by my method, but since I have not heard any more about it, I assume that my offer has been refused....

"... In your letter you ask about the pneumonia cocci and an answer has been given in the report from the congress in Berlin, where Friedländer climbed down concerning his capsules saying that he had not claimed them to be anything especially remarkable. Admittedly, there is something "fishy" about Fr.'s pneumonia cocci. His whole work was very thin, it appears to me. All his cultures and experiments are derived from 1 (one) case which in its pathological anatomy shows great difference from the ordinary pneumonias. Sections of the lungs had not been examined until I found a few small bits; it was that case with cocci (actually they are more like bacteria) which were decolorized after iodine treatment and which occurred lying free [i.e., extracellularly] in the alveoli without much exudate. In all the pneumonia cases I saw in Berlin (14-15) I only twice found a few capsulated bacteria in the dried specimens together with large amounts without capsules, and the majority of the bacteria in specimens from sections occurred inside the cells of the exudate. Also, in all the cases I observed cultivation yielded cultures of a different kind and several other bacteria gave Nagel-cultures. I believe that intrinsically the capsules have nothing to do with the bacteria but are only due to the medium in which they occur; in favor of this speaks that cultures (gelatin) never show capsules and that dried specimens—in contrast to specimens from sections—easily show capsule formation. Moreover, I believe that Friedländer now shares my view to a higher degree. At first he was very high and mighty, but at the congress in Berlin he seems to have climbed down. Now let us see this summer, I shall probably have a word to say."
I shall be at your service if you will be content with a production à la menagerie showman with traces of original comments under the title "Bemerkungen über die Färbung der Schizomy- ceten in Schnittpräparaten. (Mit Demonstration von Präparaten)," and I will send a short summary of these comments in a few days. . . ."

Letter 9, dated Strassburg, 19 July 1884

"Enclosed I am sending you some lines intended to function as the text for my demonstration of stained bugs with and without capsules. I have been somewhat puzzled as to what I ought to say; my considerations were based on the presence of Friedländer at the congress and I had in mind a little criticism, but now I cannot very well do that, so therefore my communication has become somewhat colorless."

"I hope to be able to borrow a couple of microscopes with Abbé for the demonstration, as I would be reluctant to drag my own ² to Copenhagen. . . . I have had the unexpected pleasure that in Pesti my iodine method resulted in the discovery that the pneumonia cocci are crystalline structures—tetraedres—octaedes—prisms, etc., which means that a scholar has confused the crystalline dye formed by the action of iodine upon crystal violet with the actual microorganism."

Before a discussion of what Gram did or did not understand of the significance of his staining method, it is necessary to realize that, according to present-day views, there could be an understanding at three levels: (i) its significance for the correct interpretation of the results obtained in Friedländer's laboratory at the time; (ii) its significance as a generally useful method in the day-to-day routine differentiation of bacteria, and (iii) its significance as a taxonomic criterion in the hierarchical classification of bacteria by reflecting important differences in the wall structure of procaryotes. Obviously an understanding at the third level could not be expected in 1883, so the discussion will be limited to the first two points.

Initially, it is important to note that in his publications Gram never directly indicates agreement or disagreement with Friedländer's ideas concerning the etiology of lobar pneumonia; he strictly deals with the results of his staining method and, although his results are listed in two groups under the headings I and II as, respectively, Schizomyces that are "gefärbt" (i.e. stained) and those that are "entfärbt" (i.e., decolored) by his method, he does not add any comment on the possible significance of these results for the interpretation of Friedländer's work or for that matter on a more general use of the method in bacterial differentiation.

In his letters his attitude is different; here he does not conceal that he thinks there is something wrong with Friedländer's work and directly expresses the opinion that there are at least two varieties of pneumonia cocci involved that look very different. One is stained after his method, occurs predominantly inside the pus cells of the exudate, and is found in the majority of the examined cases of lobar pneumonia. The other is decolorized after iodine treatment, it looks more like a rod than a coccus, and in the alveoli it mainly occurs outside the pus cells. This form is much rarer than the first one, but it is from a case of this kind that the cultures were obtained which Friedländer had used in his animal experiments.

About Friedländer's position the letters tell that at first he is delighted with Gram's method, but after a couple of months he asks Gram to withhold his paper temporarily because, as Gram says, it would have to contain certain disclosures—obviously the above-mentioned facts that Gram had discovered in the meantime.

As a good scientist Friedländer eventually acknowledges these facts as witnessed first by the appearance of Gram's paper on 15 March 1884 (6) containing all the essential information, but without pointing out its significance in relation to Friedländer's concepts on the etiology of lobar pneumonia. Secondly, in the discussion with Fränkel at the 3rd Congress of Internal Medicine in Berlin on 21 April 1884 (4, 5), Friedländer admits that neither capsules nor Nagel cultures are truly specific and that he considers it very likely that there are different agents producing pneumonia, although he prefers to think that in cases where cocci had been seen, but cultures were negative, the cocci had already died. As Gram says in his letter, he climbed down from his previous position.

Probably Friedländer's anticipated clash with Fränkel (Fränkel had been engaged in pneumonia studies since early in 1883, and rumors about his results had no doubt already reached Friedländer and Gram) at the Congress in Berlin about one month after the publication of Gram's paper is the reason why Gram is so careful about what he says in his first paper. In his later paper at the Congress in Copenhagen (7), Gram concludes that time may show whether it is a matter of different stages of the same pathogenic organism or whether completely different infections are under consideration.

This explanation for Gram's reticence may sound improbable to a modern ear, but these events took place almost 100 years ago when customs were different. Gram had been educated in his bourgeois home in Copenhagen to show the very best of social conduct, and now he
had been received with friendliness by Friedländer and had profited from his teaching during several months, so for him it was only natural to be as gentle as possible when he had to express opinions that were in opposition to those of his benefactor, even if it meant that he had to keep his own merits somewhat in the background. From the information in his letters I find it most probable that Gram had fully realized the significance of his method in this special issue, although his conclusion regarding the two varieties was not and could not with the limited experience at hand be based on the staining alone.

Whether Gram also recognized the usefulness of the method more generally in the identification of bacteria is more questionable; at least there is no direct evidence for such a conclusion either in his publications or in the letters to Salomonsen, and since Gram did not take up work on the staining method later in his life and later published or unpublished contributions from Gram's hand on the matter have not been found, the question must remain unanswered.

In summary, the contents of the letters lend support to the supposition that Gram had a better understanding of the significance of his method than has previously been assumed, and that the restrained account in his published articles is due to a desire not to make things worse for Friedländer in an embarrassing situation.

Notes

1 Carl Friedländer (1847-1887), pathologist at the municipal hospital Friedrichshain in Berlin, had since September 1881 studied the etiology of lobar pneumonia. Founder and editor of the journal *Fortschritte der Medizin*.

2 The content of the paper read in the Physiological Society probably corresponds to the article published on 15 November in *Fortschritte der Medizin* (3).

3 Apparently Salomonsen had asked Gram to persuade Friedländer to take part in the international medical congress in Copenhagen in August 1884 and talk about his pneumonia cocci. In the preliminary program of the congress published in May 1884, it is announced that Friedländer will demonstrate "Der Micrococcus der grupösen Pneumonie."

4 Friedländer already at that time suffered from pulmonary tuberculosis.

5 Gram's paper appeared on 15 March 1884; see reference 6.

6 Friedländer's paper from 15 November 1883; see reference 3.

7 In Koch's laboratory at the Reichsgesundheitsamt only Löffler was present. Koch and Gaffky were on their choral expedition to Egypt and India.

8 Anker Frode Rasmussen (later Halk) (1857-1927), Danish medical doctor, acquaintance of Gram and Salomonsen; studied in Berlin, 1884.

9 Carl Weigert (1845-1904), German pathologist; the first to introduce aniline dyes in bacteriology; a friend of Salomonsen.

10 Alexander Friedenreich (1849-1932), Danish medical doctor; secretary of one of the sections of the congress in Copenhagen, August 1884.

11 The 3rd German Congress of Internal Medicine held in Berlin 21-24 April 1884. The genuine pneumonia was one of the themes. Both Friedländer and A. Fränkel contributed, and here their controversy started (see reference 2 and 5).

12 Nagel cultures are nail cultures, a descriptive term used by Friedländer to characterize the appearance in sideview of a gelatin stab-culture of the organism *Klebsiella pneumoniae* he had cultivated from a case of pneumonia; the nail appearance was at first believed by F. to be of diagnostic value for this organism.

13 At the congress in Copenhagen, August 1884.

14 The final program of the congress published in August 1884 announced that Friedländer's demonstration of "Pneumonie-Kokken" would be "bei Dr. Chr. Gram." Apparently this demonstration was eventually combined with Gram's remarks on the staining of slides from tissue sections.

15 Gram's text, "Über die Färbung der Schizomyce- ten in Schnittpräparaten," is published both in the *Programme Démentif* from August 1884 and in the *Compte-Rendu* from 1886 of the Copenhagen Congress; see reference 7.

16 This Seibert microscope with Abbé condensor and an extra Leitz immersion lens (1/12) is preserved at the Medico-Historical Museum of the University of Copenhagen.

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Literature Cited


