MYOCARDIAL DEGENERATIONS IN YELLOW FEVER *

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The nature and mode of production of the toxin in yellow fever is still unexplained. Its widespread effects and influence are seen clinically and can be demonstrated pathologically, in all parts of the body. In the past particular attention has been paid to the degenerations occurring in liver, heart and kidneys. However, as yet, no satisfactory explanation has been given for the part which each of these plays in producing the death of the patient. There is ample evidence, in the clinical course of the disease, that all three of these organs are involved. Jaundice usually appears early in the course of the disease, the sclerae being jaundiced on the second or third day in many cases. The intensity of the jaundice increases with the subsidence of the fever, and may persist well into the convalescent period (Klotz,[1] Elliott,[2] Noguchi,[3] 4th Report of the West African Yellow Fever Commission[4]). As a rule the depth of the jaundice is an indication of the severity of the disease, but in many fatal cases there is little or no jaundice (Klotz).

Uremia is considered by some authors (Elliott[2] and Seidelin[5]) to be the terminal event in all fatal cases. "Albuminuria appears usually on the third day; when it appears on the first day it goes to a fatal termination; on the second day it is a very bad augury."[4] Beeuwkes[6] stated that "albuminuria was by far the most striking feature of this disease; in the Assamenkase epidemic of 1926, in all fatal cases, it was present in large amounts."

Elliott stated that when complete anuria occurs death usually follows in a few hours. In our own series, where histories were necessarily incomplete, one is struck with the frequency of the statement that "complete or almost complete anuria preceded death." Elliott noted the following as manifestations of uremia: air hunger, peculiar whistling respirations, precordial distress, headaches, backache, persistent vomiting, hiccough and finally delirium, Cheyne-Stokes respirations, convulsions and coma. That these are necessarily signs

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of uremia in yellow fever patients, who are already subjects of an intense toxemia and definite heart lesions, is a debatable point. The toxic effect of the disease upon the heart is made evident in three ways, by dilatation, weak soft heart sounds, and disturbances in pulse rate. The damage done to the heart is not purely transitory, nor is it limited to the acute phase of the illness; the heart is so weakened in some cases that death occurs when the patient is apparently convalescent (Seidelin). The disproportion between the height of the fever and the pulse rate (Faget's sign) is observed and reported in most accounts of the disease (Seidelin,[7] Noguchi,[8] Thomas,[9] Elliott, Beeuwkes, and others). This relative bradycardia may be of such extent that a patient with a temperature of 39.5°C may have a pulse rate of 80, and during convalescence a true bradycardia of 32 to 36 beats per minute may develop.

On reviewing the reports of postmortem examinations, one finds that particular attention has been paid to the gross pathological changes found in the heart in yellow fever. The microscopic pathology of this organ has been somewhat neglected, largely because the findings were so disappointingly slight in contrast to the marked macroscopic changes seen in fresh preparations (Otto and Neumann).[10] One of the best accounts of the microscopic cardiac pathology in yellow fever is given by Rocha-Lima.[11] He found that without exception there was more or less fatty degeneration of the myocardium. This was distributed in a somewhat patchy manner but was always more widespread than granular degeneration. On the other hand he seldom encountered myocardial hemorrhages. Nuclei were always well stained and often of strikingly large dimensions; the muscle fibrils and cross striations could always be clearly made out. Vacuolar degeneration, he stated, was well marked but interstitial processes were never noted.

Otto [12] reported varying amounts of fatty degeneration and vacuolar degeneration but no further changes in cells or nuclei. Otto and Neumann could not convince themselves of the fatty degeneration in the subendocardial and subpericardial regions as described by Sodré and Couto. They found that there was nothing remarkable in the muscle fibers. Both cells and nuclei stained well, and as in other organs evidence of inflammatory reactions were lacking.

Seidelin [13] noted a fatty metamorphosis of the myocardium, which was frequently less marked than the gross appearance would lead

one to expect. The aortitis and endocarditis described by Sodré and Couto were regarded as evidence of secondary infection.

Noguchi stated that the muscle fibers showed one or more vacuoles situated in the central portion, suggestive of fat. Certain fibers appeared somewhat swollen. The nuclei were large and vesicular.

Marchoux and Simond[14] noted that in the majority of cases the muscle fibers of the heart were slightly injured by the fatty degeneration, which may be made evident by scant traces of fatty granules throughout the length of certain fibers. On the other hand fatty degeneration may be very marked but was always limited to certain fibers beside which one saw others which were almost perfectly normal. Aitken, Connal, *et al*,[15] reported that in the Lagos epidemic of 1925 the cardiac muscle showed a distinctly cloudy appearance and the fibers contained small vacuoles.

Elliott gave a detailed report of the gross and microscopic pathology observed in Guayaquil, Ecuador, in 1918. He noted that subpericardial and occasionally intramyocardial hemorrhages were found. The muscle fibers were swollen, the striations indistinct, and in the most severe cases entirely absent in places. The nuclei of the most affected fibers were absent.

The pathological findings outlined above were essentially similar in character to the degenerative lesions produced in the liver and kidneys. In the former there were great varieties in the intensity of degeneration, midzonal necrosis being a characteristic feature in typical cases. Fatty deposit was variable and was commonly found in all zones, but in mild cases may be limited to the midzonal areas, and in advanced cases may be most prominent in the portal zone. The distribution and intensity of the degeneration varied in different parts of the liver.

In the kidney, too, there was always degeneration, but the amount of change was not uniform. All stages from marked cloudy swelling to necrosis of epithelial cells were found. Casts and granular débris were seen in the renal tubules. Fatty degeneration was variable, often intensely involving all the tubules of the cortex and medulla. "In general the lesions in these three organs and throughout the body are of a degenerative type, with little or no evidence of proliferative response in fatal cases, and an absence of a primary inflammation" (Klotz[16]).

Outline of Work

The material which forms the basis of this study was placed at my disposal through the kindness of Professor Oskar Klotz. It consisted of sections and blocks of material obtained at autopsy from twenty-nine cases of yellow fever which occurred in West Africa in 1925, 1926 and 1927. In addition sections were examined from ten *Macacus rhesus* monkeys, nine of which died from experimental yellow fever transmitted by direct inoculation from the blood of yellow fever patients or by bites from infected mosquitoes. The tenth monkey was killed and the sections examined as controls for the normal histology of the animal. The experimental and clinical work on these animals was done and reported upon by Drs. Stokes, Bauer and Hudson,[17] and the sections made available for me by Professor Klotz. Paraffin and frozen sections were prepared and stained by hematoxylin and eosin, and with Sudan III respectively. Some sections were specially stained for fat by scharlach R. and Nile blue sulfate stains. In three cases the bundle of His was sectioned, stained and examined in the same manner as the other tissues.

Myocardial Degenerations in Yellow Fever in Human Cases

Two constant features, fragmentation and granular degeneration of the myocardium, were observed in all sections stained with hematoxylin and eosin. The fragmentation of the muscle fibers was greatly increased in most cases in which the autopsy took place some hours after death. It is in part due to postmortem changes, although it may be evidence of the severity of the infection. Granular degeneration was found in all cases, but the amount and extent of it was not constant. It varied from very mild cloudy swelling in patchy areas, to involvement of large areas of the section in a marked granular degeneration. The severity of the degeneration in these latter cases, however, was not always more marked than that in which small patchy areas alone were involved. In mild cases the cross striations were not lost, but in more severe degenerations the fibers were swollen, pale staining and cross striations were lacking. In some sections curious variations in the staining of different fibers were evident. Patchy areas stained a deep pink, with a hyaline-like

appearance, were seen in some areas, whereas the greater number of the fibers stained a faint pink. Nuclear changes were seen in most of the cases examined, and varied in intensity with the amount of granular degeneration. They consisted of changes in the size, shape and staining qualities of nuclei. In cases where granular degeneration was extremely marked, nuclei had entirely disappeared. In some sections some nuclei were twice the size of others in the same area, and while some stained a deep and fairly uniform blue, others took a faint stain and appeared granular, pyknotic or vacuolated.

Small punctate hemorrhages were seen in only a few cases. They varied in size from small collections of five or six cells to clumps containing twenty to thirty red cells. In the cases mentioned the hemorrhages were not scattered diffusely throughout the section, but were limited to focal areas. The blood vessels were markedly engorged in two instances, but this was not a marked feature of the condition.

Inflammatory changes were noted in ten cases, but of these four were old pericardial lesions, and four consisted of thickenings of the walls of blood vessels together with small perivascular collections of mononuclear cells. In two cases, however, the inflammatory reaction was evidently in response to the intensity of the myocardial degeneration. In one of these, Adjei, the cellular exudate was made up of mononuclear cells, lymphocytes, plasma cells and endothelial cells in varying numbers. It was most evident where the granular degeneration was greatest, but was also seen perivascularly throughout the section. In the other case, McMillan, the reaction was limited to one small area, and consisted of focal collections of white blood cells, particularly polymorphonuclear leucocytes, in areas in which granular degeneration was most marked.

So-called vacuolar degeneration was seen in a few instances. This, however, was most evident in cases where the autopsy occurred some time after death. Consequently this vacuolation along with fragmentation is considered to be largely a matter of postmortem change and artefact.

In sections stained for fat it was always present. The extent and distribution of the fatty degeneration was markedly variable. In many cases it was not proportional to, or indicative of, the myocardial damage, as judged by granular degeneration, commonly being more extensive than the latter. In the majority of cases there was a patchy distribution of the fat throughout the fibers, the fatty

deposit being most marked about the nuclei. These fatty changes about the nuclei are not to be confused with the golden-brown granules of brown atrophy which are found particularly at the ends of nuclei. The fat was present in longitudinal rows of fine granules, throughout the length of the fibers, increasing in size to small droplets in the neighborhood of nuclei. In milder cases the fatty degeneration was seen only about the nuclei, and its patchy distribution in the section was markedly emphasized. In severe cases fatty degeneration was very marked, practically all fibers being affected, but even here the patchy distribution of the lesion was noticeable, some areas showing considerably more degeneration than others.

Unsatisfactory results were obtained in sections stained by Nile blue sulfate, since the fat appeared sometimes red, sometimes blue, and all gradations between these two colors were observed. One cannot therefore draw any conclusions from this study as to the chemical nature of the fat.

On examining sections taken from the conducting bundle the results were disappointing. In one case, Mrs. Elmore, there was a marked fatty degeneration of the bundle. In the two other cases no fatty degeneration was observed. One cannot conclude from these findings, however, that fatty degeneration of the conducting bundle was not present in more than one third of the cases. There was only a slight fatty degeneration present in sections of the myocardium from these two cases. We have found that this was not an infrequent occurrence in specimens preserved for long periods in formalin. This was particularly evident in one case where sections were stained immediately postmortem, six months later, and then again eighteen months later. The fat was markedly decreased in the second instance, while in the third set it had almost entirely disappeared. This, however, was not always the case; some specimens seemed peculiarly susceptible to the action of formalin. In so far as could be determined the formalin was the same in all cases.

Summary

Fatty degeneration was observed in all cases. The distribution and intensity of the degeneration varied from field to field in the section, and from point to point in the fibers, being particularly concentrated about the nuclei. The nature of the chemical composition of the fat could not be determined by the use of Nile blue sulfate

stain. The results of sectioning the connecting bundle were inconclusive; further work should be done upon both these points.

Cloudy swelling was a constant feature, but here, too, variations in intensity were marked. Nuclear changes and fragmentation of muscle fibers were features of all cases. Hemorrhages occurred in seven cases, and in these instances were very fine punctate ones. Engorgement of blood vessels was not a marked feature, occurring only in two cases. Previous inflammatory changes were present in eight cases, while two showed inflammatory exudates which had responded to very acute degenerations of myocardial fibers.

Myocardial Degenerations in the Macacus Rhesus in Yellow Fever

The experimental work upon these animals was done by the late Dr. Adrian Stokes, and by Drs. Bauer and Hudson at Lagos, West Africa. A preliminary report of their work is found in the *Journal of the American Medical Association*,[17] and a fuller report in the *American Journal of Tropical Medicine*.[18] The first animal was inoculated directly from a human case of yellow fever, the second and third by transfer inoculations from the first, and the remainder by bites from infected mosquitoes. The clinical course of the disease was similar to that of yellow fever in man, fever occurring upon the third or fourth day following inoculation. Death resulted in from one to seven days after the initial fever. The gross pathology was similar to that of human cases, and the authors were convinced that they had successfully transmitted yellow fever to the *Macacus rhesus*.

The normal microscopic anatomy of the heart of the *Macacus rhesus* differed considerably from that of the human. The myocardial fibers were narrow and closely compacted, cross striations were well marked and easily made out. The nuclei were variable in size and shape but were usually rounded and oval in form, and large in comparison with the size of the fibers. The walls of the blood vessels were thickened. There was also an increased amount of connective tissue with a slight exudate of lymphocytes and endothelial cells about them indicative of previous inflammatory changes. Autopsies were performed immediately after death, the animals either dying of the disease or being killed by a whiff of chloroform while in a moribund state.

Sections were made from paraffin and frozen blocks of tissue. These were stained routinely by hematoxylin and eosin, and Sudan III respectively.

In contrast to human cases fragmentation was not a marked feature. However, cloudy swelling and granular degeneration were present. They varied in amount and distribution, but in severe cases cross striations were obliterated. In some sections there were patchy areas which stained markedly acidophilic, the muscle substances appearing very much like hyaline material. Nuclear changes were present in most cases. The extent of these changes, however, was not so marked as in human cases, variability in the size and staining of the nuclei was observed in normal hearts. Occasional punctate hemorrhages were seen. Inflammatory changes, aside from those seen in the normal heart, were not observed, and vacuolar degeneration was missing.

Fatty degeneration was present in all the cases studied. Its extent was variable and ranged from that seen in one case, in which only a minute quantity was present about the nuclei, to that in others in which the whole section was heavily loaded with fat. The patchy distribution of the degeneration was quite marked. The fat was found more particularly in the region of the nuclei. In instances where it was distributed throughout the fiber it did not have the regular linear arrangement of granules seen in human cases. The fat appeared in the form of fine granules, becoming somewhat more globular about the nuclei.

Summary

Fatty degeneration was a constant feature varying in severity from very slight fatty change to a marked general degeneration. The patchy distribution of the lesion was seen characteristically whether the case was severe or light. Cloudy swelling and granular degeneration were constantly present, but here, too, variations in the intensity of degeneration, and patchy distribution of lesions, were observed. Nuclear changes were present in most cases. Hemorrhages were not a marked feature of the condition, occurring only in two cases. Capillaries containing red blood cells were evident in large numbers in two cases. Inflammatory changes were seen in seven cases, but these were not in relation to the disease and were

not more marked than in the normal animal. Patchy areas of hyaline-like degeneration were seen in three cases. Fragmentation and so-called vacuolar degeneration were absent in these cases.

Comparison of Findings in Macacus Rhesus and Human Cases

The pathological findings in the heart were essentially the same in the human and in the *Macacus rhesus*. Cloudy swelling, granular degeneration, fatty degeneration and nuclear changes were common to both. The intensity of the fatty degeneration was somewhat less marked in the *Macacus rhesus*, whereas its patchy distribution was intensified. The distribution and arrangement of the fat was not so regular here as in human cases, but was similar in that it was most marked about the nuclei. Hyaline-like degeneration was a feature of the changes in the myocardium of the monkeys which seemed to be more marked than in human cases. In no case was the degeneration sufficiently intense to produce an active response of leucocytes such as was seen in two human cases. Fragmentation and vacuolar degeneration were absent in the monkeys. This further strengthens the view that they were largely due to postmortem changes, rather than results of the yellow fever, since these animals were autopsied immediately after death.

Owing to the difficulties encountered in obtaining histories and the late period at which natives seek medical aid, the clinical data in many of these cases are necessarily somewhat incomplete. Eighteen cases occurred in whites, eleven in negroes. Twenty-four cases were males, five females. The average age was 32, the youngest 4 and the oldest 57 years. Two cases occurred in children aged 4 and 5 years respectively. The onset in most cases was sudden and severe, with chills, fever, nausea and vomiting, headaches and pain in the loins. The clinical course of the disease was usually rapid, death occurring in four or five days in the majority of cases, the longest being nine days and the shortest two. Fever, black vomit, melena, anuria, jaundice and slow pulse were outstanding in the clinical signs. Jaundice was usually not very marked before death, but at autopsy after the congestion of the skin had subsided it was made out fairly well. However, in a surprisingly large number of cases, sixteen to be exact, it was noted as being very slight, while in some five cases, only mild

or moderate scleral jaundice was noted. Fever was not excessively high in any case, ranging from 100° F to 102° F in most cases and reaching a maximum of 104° F in one case. Faget's sign, or the disproportion between the pulse rate and temperature, was evident in all cases where these data were taken. This was in keeping with the findings of other observers. The causation of this slow pulse rate has been generally explained by the presence of bile salts in the blood stream. King and Stewart [19] noted that the amount of bile salts in a dose of pig's bile lethal for dogs, if injected alone, will produce neither fall in blood pressure nor slowing in rate. However, they found that the amount of bile pigment contained in a dose of pig's bile lethal for dogs will if injected alone cause death, with slowing of the heart and falling blood pressure. They believed the bradycardia to be a direct result of heightened vagus tone (produced by the action of bile pigments) as atropine restored the rate. They also observed a delay in conduction time between the auricle and ventricle amounting to 2/100 to 5/100 seconds.

More recently, doubt has been cast upon the relation of jaundice and bradycardia. McVicar and Fitts [20] have stated that "bradycardia in jaundice has in our experience proved almost a myth; when it has been observed it has given no clue to the diagnosis." An editorial of the *Journal of the American Medical Association* [21] stated that bradycardia was an infrequent accompaniment of jaundice, except in the intrahepatic type or the so-called acute catarrhal jaundice. Clinically in three cases that have come to our own attention recently, where jaundice and fever had been present, slow pulse was not observed. The first case was that of a young male aged 24 with an unexplained jaundice which was marked in the sclera, palms of the hands and soles of the feet. The temperature was 100° F to 101° F and persisted for five days, the pulse rate varied from 90 to 110 during the fever and subsided with it to the normal rate of 74 to 80. The second case was that of a woman dying of eclampsia with marked jaundice, some fever and a pulse rate of 110 to 120. The last case was that of a young woman aged 28, with subacute yellow atrophy; jaundice was intense (duration seven to ten days), temperature 100° F to 101° F and pulse rate 134. As a consequence of these findings, and the fact that in many fatal cases of yellow fever jaundice was not marked, one must conclude that the slow pulse rate here was not due to jaundice. The finding of marked

fatty degeneration in the bundle of His in one case was suggestive and may be a possible explanation of the slow pulse in yellow fever. Further clinical and pathological studies should be done in order to confirm or disprove this.

The findings in the above series of cases were, in general, in harmony with those previously reported. The presence of inflammatory cells in the heart in response to acute degeneration has not been noted before, although it has been reported in the liver by Klotz. The intensity of the myocardial degenerations occurring in the heart in yellow fever was in itself sufficient to account for death in some cases; but in others it must be taken as only contributary in producing death, which was induced by the general toxemic effect of the disease upon the whole body, particularly heart, liver and kidneys. The pathological findings in the heart were not sufficient to make a diagnosis of yellow fever, but taken together with the changes observed in liver and kidney are satisfactory evidence upon which the diagnosis may be made. The observations recorded in the hearts of *Macacus rhesus* were quite similar to those seen in human cases. These findings taken in conjunction with those in other organs make it apparent that a susceptible animal has been found for yellow fever. Noguchi [22] reported that the *Macacus rhesus* was resistant to leptospira icteroides strains isolated from yellow fever cases in Guayaquil, while marmosets succumbed to the infection with pronounced symptoms. Their findings in these animals were different from those in our report. In hematoxylin and eosin sections the striations were visible but slightly less distinct than normal. The muscle fibers were the seat of numerous very minute vacuoles. Nuclei were normal and no hemorrhages or other forms of degeneration were present. In scharlach R. sections very fine fat droplets were sprinkled uniformly throughout the entire length of all the muscle fibers. There was no accumulation of fat about the nuclei. Similarly Muller [23] reported that there were no hemorrhages or any form of degeneration such as Zenker's. In scharlach R. sections (one monkey alone was examined) numerous fine fat droplets were sprinkeld uniformly throughout the entire length of all muscle fibers. The fact that these findings were at variance not only with those in human cases, but also with those found experimentally in our series of *Macacus rhesus* suggests that the infection under these different conditions was not the same.

Summary and Conclusions

1. The microscopic examination and analysis of the hearts of twenty-nine cases of West African yellow fever, and those of nine monkeys experimentally infected with West African yellow fever, is here reported.

2. Cloudy swelling, granular and fatty degeneration were found constantly in the hearts of both the human cases and those experimentally induced in the *Macacus rhesus*.

3. Primary inflammatory changes were not seen in the heart in yellow fever. Secondary response of white blood cells to intense degeneration was observed in two human cases.

4. The distribution and intensity of granular and fatty degeneration was patchy and variable in both human cases and *Macacus rhesus*. Fatty degeneration was most marked in the neighborhood of nuclei of the fibers.

5. The causation of the slow pulse in yellow fever is still uncertain, and doubt is thrown upon the belief that it is due to the jaundice.

6. Further investigation of the clinical function and the pathological changes in the bundle of His may lead to solution of the problem.

7. The lesions in human hearts and in those of the *Macacus rhesus* are essentially the same.

8. The lesions in the heart are in themselves not sufficient to make a diagnosis of yellow fever.

In conclusion I wish to thank Professor Oskar Klotz, at whose suggestion this work was undertaken, and who has greatly assisted me during its prosecution by advice and direction.

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