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# Three Stages of *Cholera*

**Toyosuke WATANABE, Takeshi SHOZAWA, Yukie KUNITA and Ichiro SEKINE**

*Department of Pathology*

**Kazumine KOBARI, Setsuko SOGAME and Masao NAKATOMI**

*Department of Clinical Medicine*
*Institute for Tropical Medicine, Nagasaki University*

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**Abstract**

Cholera is regarded as the most acute of the acute enteritides.

This fact has apparently caused the general impression that there is only one stage in the course of cholera. So acute as it may be, a clear cut shift of pathological changes in the intestinal mucosa of cholera patients are recognizable, if we arrange autopsy cases in time order after onset of the disease. There are 3 stages, the first, stage of inflammation, the second, stage of villus fusing and the third, stage of villus resolution. The materials were 12 cases of infant cholera, which died in 1968 and 1969 in the San Lazaro Hospital, Manila, Philippines.

Since the work of Gangarosa,[1] "The nature of the gastrointestinal lesion in asiatic cholera and its relation to pathogenesis" was reported, no important publications have been made for 10 years about the pathological anatomy of cholera. The work of Gangarosa was so epoch-making, that most of the pathologists seem to have stopped studying about pathology of cholera. They must have thought, that the pathological study of cholera has been completed, so far as the human morphology is concerned. Only some supplementary works have been reported aiming at confirmation of the Gangarosa's conception, "Epithelial lining is intact in cholera."

Does the work of Gangarosa really mean an completion of the cholera pathology ? No, by no means. The importance of his achievement consists in the decisive negation of Virchow's denudation theory, which has impeded over 100 years the development of cholera pathology, but not in the completion. On the contrary, his great contribution to the cholera research is in that he has set free cholera patholgy from the wrong preoccupation and given the green light to the pathologists. It is quite regrettable that most of the pathologists mistook the green signal, "Start ! ", for the red signal, "Stop ! It's completed." Those, who read Gangarosa's

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report carefully, will easily find that he remarks nothing about the relationship between tremendous diarrhea of cholera and its morphological findings.

It was reported by the author in the book, "Enteritis"[2] and at the general assembly of Japanese Pathological Society[3] in autumn, 1968, that the copious diarhea in cholera is caused by integration of hypersecretion of digestive juice, morphological changes of intestinal mucosa (such as blunting and fusing of villi, degeneration of epithelial cells) and disturbance of villus contraction. However, at that time, only a few autopsy cases of cholera were available for us. Those were not sufficient to describe the pathological shift from initial changes to recovery in the intestinal mucosa. Since then, we were able to collect almost 10 quite fresh cases postmortem within 90 min., which have yielded as good histological specimens as those obtained by biopsy. The purpose of the present paper is to show that 3 stages of pathological process is recognizable even in cholera, the most acute of the acute enteritides, when we arrange these cases in time order from the onset of the disease.

## Materials and Methods.

Twelve infant cholera cases, who died of cholera El Tor 1968 and 1969 in the San Lazaro Hospital, Manila, Philippines, were included in this study. The clinical diagnosis was bacteriologically confirmed in each case except A. N. 97, 95 and 96, of which rectal swab on admission gave negative culture of cholera vibrio, ispite of their signs and symptoms of classical cholera.

The table, "Autopsy Findings of Infant Cholera 1968 and 1969 in Manila," summarizes the necessary data, postmortem hours, clinical course, bacteriology, and histology. Nine cases were autopsied within 1hr 30min, 5 out of them within 1hr. Three other cases were autopsied postmortem 1hr 45min, 2hrs 40min and 4hrs respectively. The last one can be used only for limited purposes be-

Autopsy Findings of Infant Cholera 1968 and 1969 in Manila (Small-intestinal Mucosa)

| A.N. | Age | postmort | Course | Vibrio | Cellinfilt | Fusing | Vill./Cryp. | Epith. Desquam. |
|---|---|---|---|---|---|---|---|---|
| 38 | 6y ♀ | 4.0hrs | Some hrs | (+) | ++ | ? | 2.5 | ++ |
| 88 | 3y ♀ | 45m | 7hrs | (+) | ++ | − | 2.5 | ++ |
| 90 | 4y ♂ | 35m | 12hrs | (+) | ++ | − | 2.0 | ++ |
| 94 | 3y ♂ | 1h 30m | 13hrs | (+) | ++ | − | 2.5 | |
| 93 | 6m ♂ | 55m | 14hrs? | (+) | ++ | ++ | 1.0 | ++ |
| 97 | 2y8m ♂ | 50m | 24hrs | (−) | + | ++ | 0.4 | + |
| 89 | 3y ♀ | 2h 40m | 32hrs | (+) | + | + | 0.8 | + |
| 92 | 4y ♂ | 1h 20m | 33hrs | (+) | + | − | 2.0 | ++ |
| 95 | 8y ♀ | 60m | 40hrs | (−) | + | ++ | 0.8 | ++ |
| 96 | 5y ♀ | 1h 05m | 40hrs | (−) | ++ | ++ | 0.4 | + |
| 91 | 1y7m ♂ | 1h 05m | 41hrs | (+) | + | ++ | 0.4 | + |
| 39 | 2y ♀ | 1h 45m | 7ds | (+) | + | ++ | 0.4 | ++ |

cause of its advanced autolysis of intestinal mucosa. In order to minimize autolytical changes, the intestine was first taken out from the cadaver and opened at once, 2 pieces each were taken from jejunum, ileum and colon and fixed immediately in Zenker Formol fixative, contrary to the orthodox autopsy technic, which needs 1-2hrs until opening the intestine. In the following chapter, we treat, however, only the small intestine, especially jejunum, for, as is discussed in the author's previous work, the colon is nothing but a byplayer in the diarrhea.

## Results

As is seen from the table, inflammatory cell infiltration in the small-intestinal mucosa is marked in early cases, some hours, 7, 12, 13 hours after the onset of the disease manifested by diarrhea and/or vomitting, whereas fusing of villi is completely absent. We want to call this stage, "stage of inflammation". However, it must be here emphasized, that the intensity of cholera enteritis, characterized by catarrhal enteritis, is less than that of other forms of enteritis, such as erosive, ulcerative or pseudomembranous enteritis. Prodigious diarrhea in cholera occurs, because its enteritis is slight, in other words, because the digestive juice is well secreted due to lack of circulatory disturbances.

In over 24 hours, blunting and fusing of villi dominate the histological scene resulting in tremendous decrease of surface area, while inflammation in the lamina propria is subsiding.

The III stage, called by the author the stage of villus resolution, is still an theoretical one. In A. N. 39, 7 day after onset of disease, the fusing is still marked. Perhaps it will need 2 or 3 weeks, until fused villi are again separated.

In gross estimation, villus height-crypt depth ratio is high in the I stage, as compared with the II stage, though it is already lower than that in normal intestinal mucosa.

As for the epithelial desquamation, it is clear from the table that the desquamation is quite pronounced in the stage of inflammation, though this finding is influenced by several factors, first by postmortem hours as in A. N. 38, then by the amount and enzym activity of digestive juice in the intestinal lumen and last but not least by the autopsy technics including fixation method.

## Histology

Fig. I. A. N. 38. H. E. x750. Postm. 4hrs. Some hours after onset of disease. Jejunum.

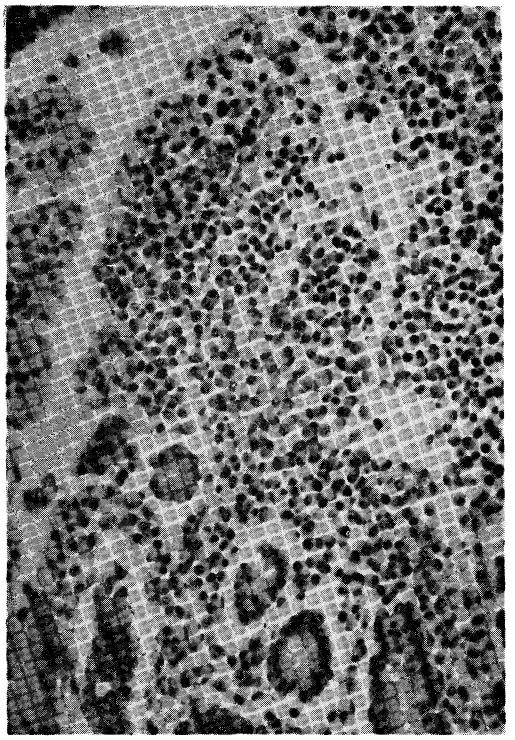

**Fig. I**

The villi are completely denuded of epithlial linings because of long postmortem hours. Lamina propria is widened by strong inflammatory cell infiltration. However, there is neither marked hyperemia, nor erosion nor necrosis.

Fig. II. Normal villi (Adult case) H. E. x750. Postm. 50min. Jejunum.

Note the high epithelial lining and slender lamina propria without cell infiltration.

Fig. III. A. N. 94. H. E. x750. Postm. 1hr 30min. 13 hours after onset of disease. Jejunum.

Fig. IV. A. N. 94. H. E. x300. Jejunum.

Epithelial denudation is here and there found. An abundant mixture of mucus, leucocytes and detached epithelial cells is seen in

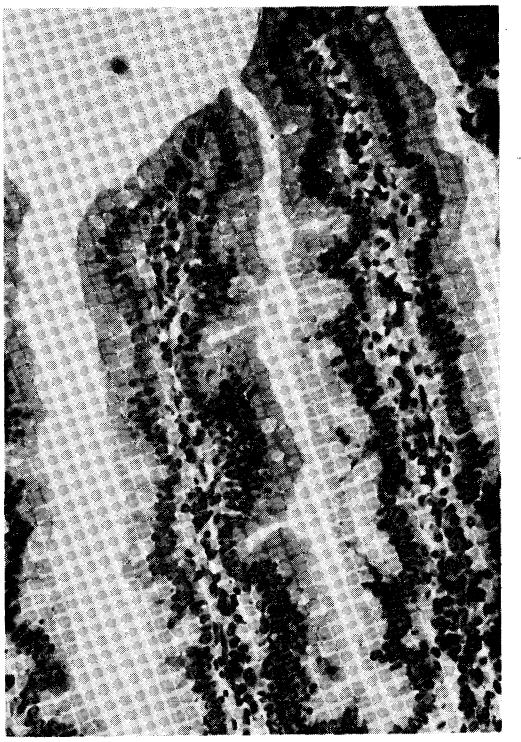

**Fig. II**

**Fig. III**

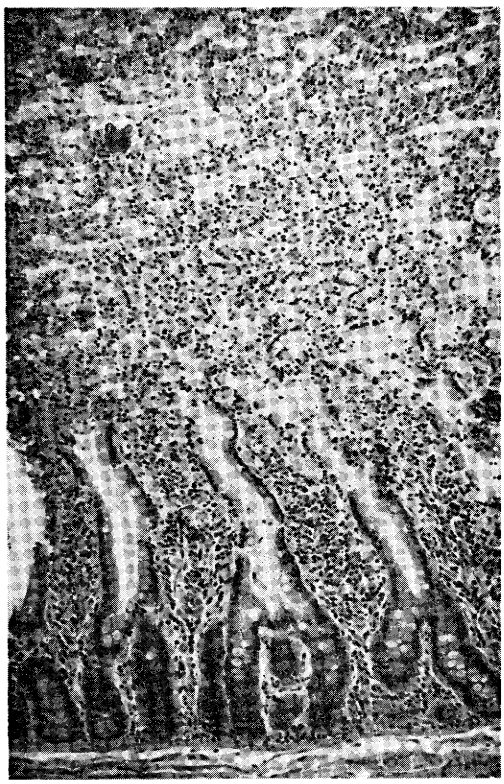

**Fig. IV**

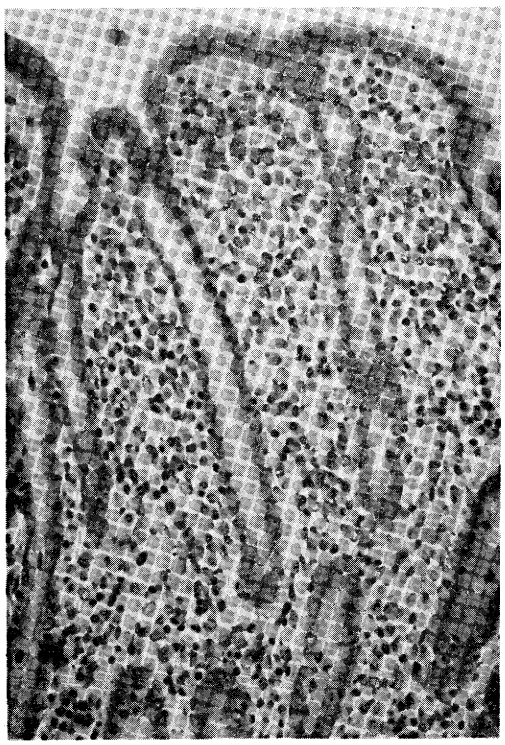

**Fig. V**

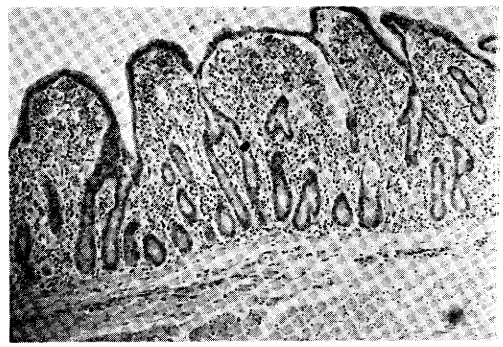

**Fig. VI**

the upper half of the figure. Cell infiltration in lamina propria is also conspicuous, whereas no fusing at all is seen in the villi. There is also neither marked hyperemia, nor erosion nor necrosis as in Fig. I.

Fig. V. A. N. 97. H. E. x750. Postm. 50min. 24hrs after onset of disease. Jejunum.

Fig. VI. A. N. 97. H. E. x300. Ileum.

Cell infiltration is subsiding, though histiocytes are still numerous. On the other side, fusing is developped everywhere, in the whole

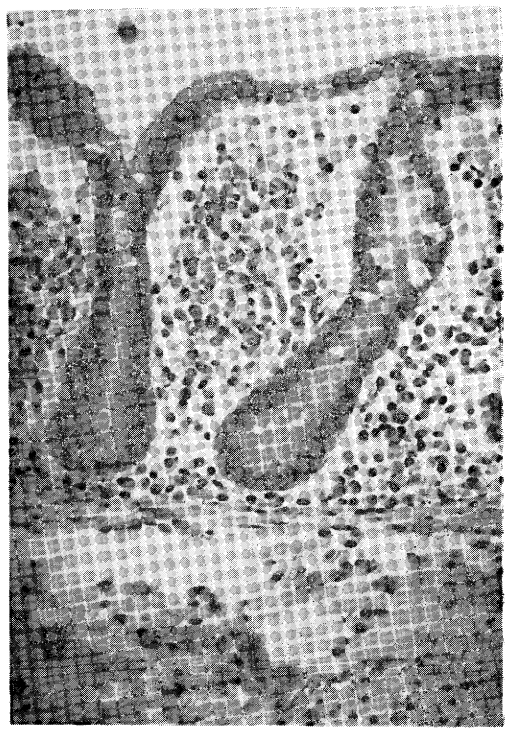

**Fig. VII**

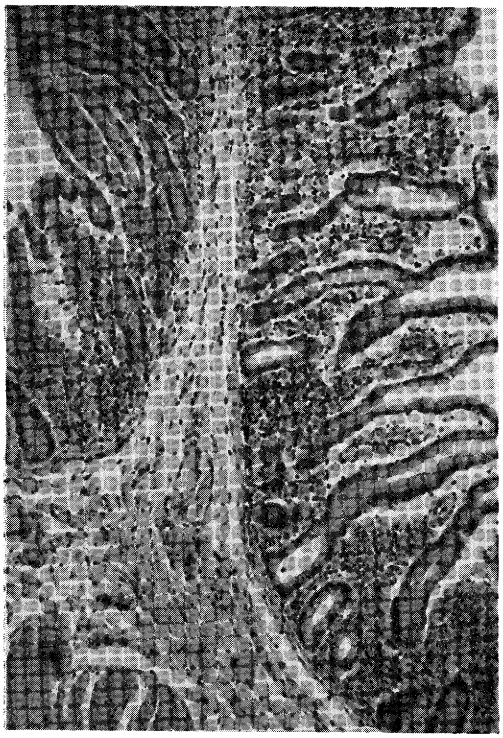

**Fig. VIII**

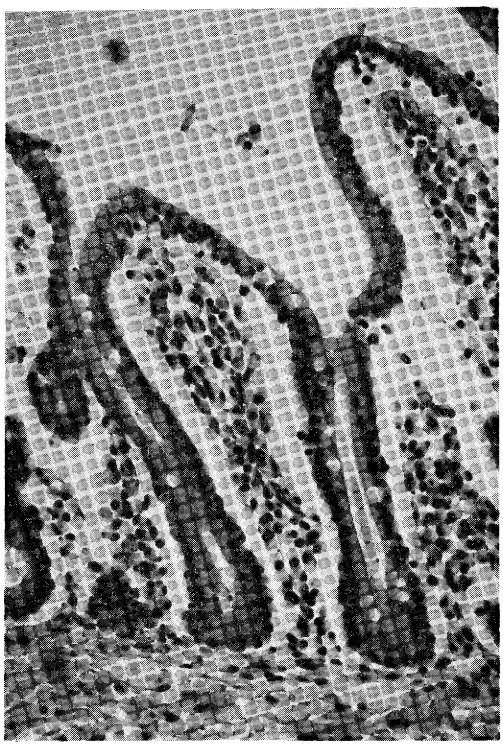

**Fig. IX**

small intestine, especially in ileum. The fusing may occur in the entire length of villi or in some portion of them, consisting in mutual adhesion of epithelial linings deprived of microvilli. Because the acute fusing is epithelial, but not proprial, it is not well comfirmable in cases autopsied over 2hrs after the death as in A.N. 38(See the table!), in which most of epithelial linings are auto lytically detached.

Fig. VII. A. N. 96. H. E. x750. Postm. 65-min. 40hrs after onset of disease. Ileum.

There is only moderate cell infiltration in the lamina propria, histiocytes predominating, and the fusing is marked.

Fig. VIII. A. N. 91. H. E. x300. Postm. 65-min. 41hrs after onset of disease. Jejunum.

Only a slight cell infiltration is seen, while fusing is fairly marked.

Fig. IX. A. N. 39. H. E. x750. Postm. 1hr 45min. 7 days after onset of disease. Jejunum.

Cell infiltration has subsided, though the fusing still remains.

## Discussion

Cholera is the most acute of the acute enteritides. This fact has apparently brought about the general impression that the pathological process in the small intestinal mucosa of cholera victims presents only one stage in its short course of illness, that is, the stage of inflammation. So acute as it may be, the pathological changes could be divided into 3 stages, i. e. the I stage of inflammation, the II of villus fusing and the III of villus resolution according to our autopsy cases arranged in time order from the onset of dis-

ease manifested by vomitting and diarrhea.

The fusing has hitherto been regarded as chronic finding. Thus, most of gastroenterologists are of the opinion, that the fusing very often observed in cholera patients indicates chronic enteritis and that the cholera affects only the persons suffering from malnutrition due to chronic enteritis. According to the author, however, there are two sorts of fusing, acute and chronic one. And fusing of villi, demonstrated in this paper, presents undoubtedly the acute one from the following reasons.

a) Fusing in our cases is epithelial adhesion, but not proprial fusion.

b) It is not observed in early cases. That is to say, it is not found before 24 hours after the onset of disease.

c) No relationship exists between malnutrition and fusing. A. N. 94 suffered from malnutrition without any fusing of villi. A. N. 97 showed marked fusing, though the mutritional condition was very good. A. N. 96, however, demonstrated malnutrition and at the same time marked fusing.

d) Fusing is rather related to the time after the onset of disease, as described above.

e) Contrary to chronic fusing, in which always blunting and fusing of villi go hand in hand, there are occasional fusings in cholera victims not accompanied by any blunting, as in Fig. X. (A. N. 97 H. E. x300. Postm. 50min. Jejunum).

f) Fusing is readily produced experimentally in guinea pigs within 24 hours.[3]

Thus, there is sufficient reason for the existence of the II stage, the stage of villus fusing. In this connection, there is often observeed a interesting finding in early cases such as in Fig. XI.(A. N. 90. x300 Postm. 35min. 12hrs after onset of disease. Jejunum,)

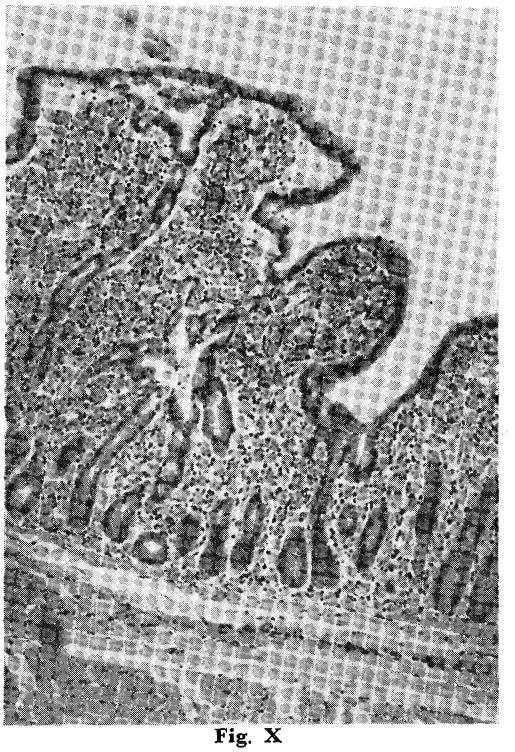

**Fig. X**

i. e. Pseudofusing, a fusing through abundant mucus serving as binding agent, which has the same functional effect as the real fusing, that is disturbance of water reabsorption.

As for the disturbance of reabsorption, there is an another interesting finding. That is the covering of almost the entire length of intestinal mucosa with a mixture of mucus, inflammatory and epithelial cells detached, which will have obviously great effects on reabsorption of water. Or we might say, water absorption is virtually ceased. Such finding is seen in Fig. IV. and Fig. XII. (A. N. 94. H. E. x120. Jejunum).

The III stage, the stage of villus resolution is at present still an imaginative one. However, we are convinced of its existence.

Otherwise, cholera patients would have to suffer from ever lasting diarrhea or malnutrition even after full recovery from the condition.

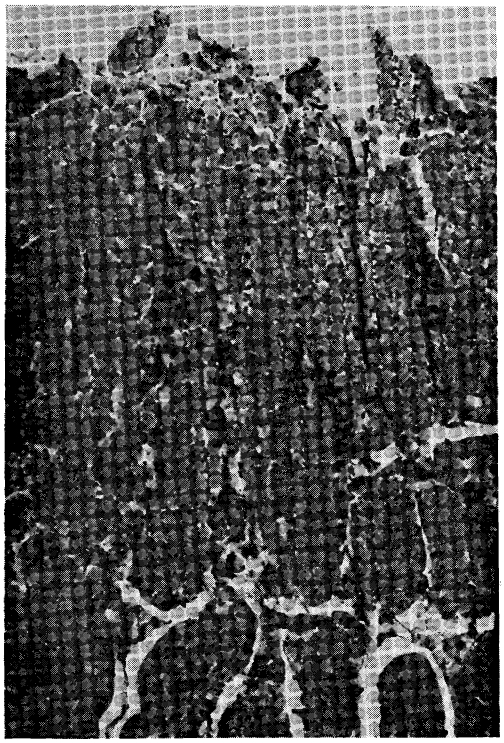

Fig. XI

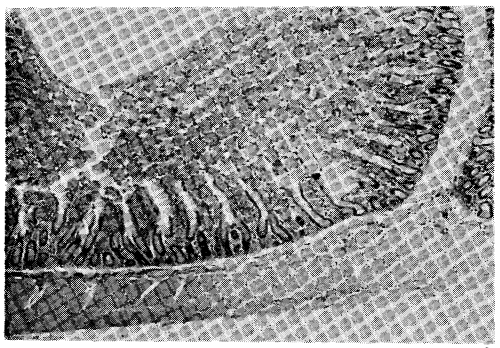

Fig. XII

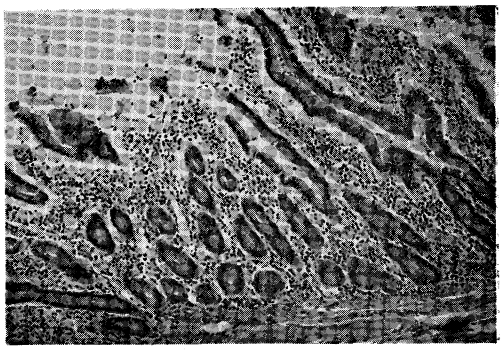

Fig. XIII

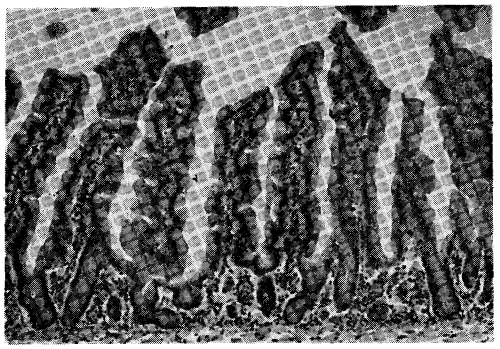

Fig. XIV

At last, we would like to discuss about epithelial detachment of small intestinal mucosa. As is seen in the table, epithelial desquamation is prominent in the early cases with strong cell infiltration, while in the late cases, it is much more silght, especially in the area with fused villi. In Fig. XIII (A. N. 88. H. E. x300. Postm. 45min. 7 hours after onset of disease. Jejunum. ), conspicuous detachment of epithelial lining is seen at the top of villi despite the short postmortem time, 45 minutes, whereas in Fig. X with marked fusing, no detachment is noticed at least in this part of jejunum. Compare with normal case, Fig. XIV, (H. E. x300 Postm. 50min. Jejunum). Supposedly, there would be no desquamation in the entire length of samll intestine of the late cases, when observed in living state, i. e. without autolysis, by biopsy, as Gangarosa has stated in his report. However, it still remains questionable, whether no detachment of epithelial lining occurs in the early phase of cholera, 10 hours or so after onset of disease, for no examination has been ever made in this phase by biopsy technics.

## Summary

1. The real start of cholera study has been made only 10 years ago. It is most urgent to collect as many fresh autopsy cases within one hour after death as possible in order to clarify the pathology of cholera.
2. Three stages are recognizable even in the short course of cholera, i. e. the stage of inflammation, that of villus fusing and that of villus resolution.
3. The III stage is still an imaginative one, though its existence is theoretically established. Further studies by means of biopsy technics are required.
4. Whether no detachment of epithelial lining occurs even in the beginning of cholera, still remains open. Biopsy studies in this stage are also needed for the solution of this problem.
5. The general conception, that the fusing observed in cholera patients is chronic, is nothing but a superstition lacking scientific proofs.



## Acknowledgement

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# コレラの三期


**渡辺 豊輔, 所沢 剛, 国田 五重, 関根 一郎**

長崎大学熱帯医学研究所病理学部

**小張 一峰, 十亀 節子, 中富 昌夫**

長崎大学熱帯医学研究所臨床部



## 摘 要

コレラは急性腸炎の中でも最も急性のものとされている．此の事実がコレラには一期しかないと云ふ観念を一般に植え付けたものと思われる．然し，如何に急性の疾患とは云え，コレラの剖検例を発病後の経過時間によって整理して見ると，小腸粘膜の形態学的変化に明かな推移が認められる．我々は此を三期に分類した．

第一期，炎症期，発病後十数時間迄．第二期，絨毛癒着期，発病後二十四時間以後．第三期，癒着緩解期，発病後推定二，三週，症例は1968年，1969年に比島，マニラ市，サンラザラロ病院で死亡した小児コレラ12例である．