

The histopathology of the possible cause and the local reaction in cobra bite

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ประไพ พงษ์ประสิทธิ์, ชูลี มิตรกุล, นกมล นพคุณ. พยาธิสภาพและสาเหตุ
ที่น่าเป็นไปได้ของแผลงูกกัด. จุฬาลงกรณ์เวชสาร 2527 ตุลาคม ; 28 (10) :
1109-1116

ได้ศึกษาพยาธิสภาพจากรอยเขี้ยวงูกัดในเด็กที่มีอาการทางประสาท และได้รับการรักษาด้วยเซรุ่มแก้พิษงูเห่า 36 ราย โดยตัดชิ้นเนื้อจากรอยเขี้ยวหลังกัดในเวลาต่าง ๆ กัน ส่วนใหญ่ภายใน 24 ชั่วโมง เพาะเชื้อแบคทีเรียทั้งชนิดแอโรโรบิก และแอนแอโรโรบิก จากน้ำเหลืองในแผลที่ตัดชิ้นเนื้อทันที 18 ราย และศึกษาปฏิกิริยาทางอิมมูนจากชิ้นเนื้อโดยวิธี *direct immunofluorescent stain* 4 ราย ผลพบว่าพยาธิสภาพมีการเน่าตายของเซลล์ผิวหนังชั้นบนเป็นหย่อม ๆ มีการเสื่อมสภาพของต่อมเหงื่อและเนื้อเยื่อคอลลาเจนเป็นหย่อม ๆ มีการอักเสบของหลอดเลือดทั้งส่วนบนและล่างของผิวหนัง รวมทั้งมีการอุดตันของหลอดเลือด พบว่าผลการเปลี่ยนแปลงทางพยาธิสภาพของชิ้นเนื้อใน 24 ชั่วโมงหลังงูกัดไม่ขึ้นอยู่กับเวลาเร็วหรือช้าหลังกัด แต่หลังจาก 24 ชั่วโมงแล้ว พบมีปฏิกิริยาเน่าตายของเซลล์ของผิวหนังทั้งชั้นบนชั้นล่าง และไขมันใต้ผิวหนัง

ผลทางแบคทีเรียส่วนใหญ่เป็นแกรมลบกลุ่ม *Proteus* ซึ่งมีพิษรุนแรง พบ *Staphylococcus aureus* และ *nonhemolytic streptococcus* ขึ้นอย่างละหนึ่งราย ส่วนเชื้อแอนแอโรโรบิก พบ *Peptostreptococci* และกลุ่ม *Clostridium* รวมทั้ง *Clostridium tetani*

สำหรับผลค่านปฏิกิริยาทางอิมมูนยังทำได้จำนวนน้อย ผลทางพยาธิสภาพของแผลงูกัดอาจสรุปได้ว่าเป็นผลจากปฏิกิริยาของพิษงู การอักเสบจากเชื้อแบคทีเรียหรืออาจเป็นผลจากเซรุ่มของพิษงูซึ่งควรจะศึกษาต่อไป

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Although Cobra is one of the common poisonous snakes found throughout Thailand only 2 of the 12 species of the Elipidae family inhabit here. They are *Ophiophagus hannah* (King Cobra) and *Naja naja*.^(1,2,3) Their toxins composed of neurotoxin, cardiotoxin, enzymes and proteins.^(2,4,5) The victims may die from respiratory muscle paralysis due to the neuromuscular blocking effect, curare-like mechanism of the neurotoxin.^(2,5) Cobra antivenom can antagonize the neurotoxin.^(4,6)

The cobra antivenom and assisted ventilation are life saving in many cases

of respiratory paralysis. The local reaction of the bite is also a serious problem in children, though not life threatening. All cases have variable swelling, pain, bruising and hemorrhagic blebs. Severe necrosis of the skin and subcutaneous tissue is seen in most cases.⁽⁷⁾ The healing process needs at least 1-2 months. Some severe cases even require skin grafting. There is no definite opinion nor previous report on the pathogenesis of the local reaction. Our objective is to report the histopathological findings and speculate about the possible causes of the local reaction.

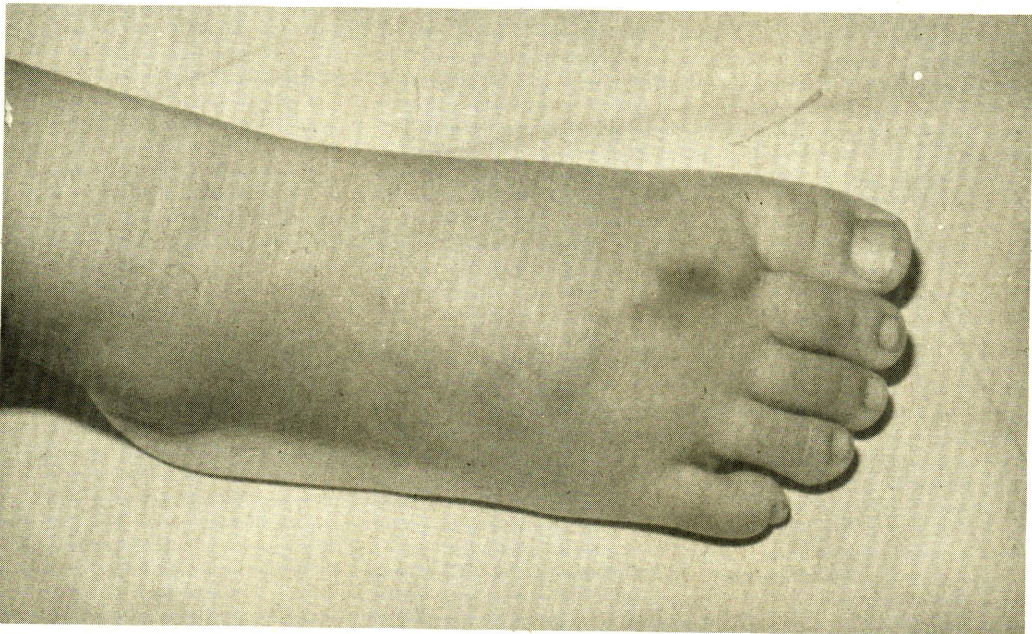


Fig. 1 Local swelling, bruising with 2 fang marks of cobra bite on the dorsum of leg (10 hr. after bite)



Fig. 2 When slough was excised 7 days after the bite at the ankle, a necrosis deep into the muscle was found requiring skin grafting. The healing process needed almost 2 months.

Materials and methods

The study was carried out at the Pediatric Department of Chulalongkorn Hospital between 1978-1983 in 36 proven cobra bite children who developed neurological manifestations and were admitted to the pediatric intensive care unit under the following criteria :

- The patient gave a definite history of cobra bite or brought the cobra with him.
- The character of cobra bite : 2 fang marks about 1-1.5 cm. apart were found by physical examination.
- Symptoms and signs of local reaction to cobra bite such as a painful rash, swelling, bruise or blistering at the bite site immediately to 48 hours after bite.^(1,7)

- Systemic neurological symptoms and signs such as generalized weakness, sleepiness from 20 to 30 minutes after bite, salivation, stiffness of tongue, difficulty in swallowing, vomiting and dysarthria 30 to 50 minutes after bite, paralysis, difficulty in breathing, inability to speak and apnea.

These patients were treated by the following regime ;

1. Give tetanus antitoxin or tetanus toxoid 1 ml intramuscularly and closely observe the patient.

2. Give cobra antivenom intravenously 30 ml every 10 to 15 minutes until the patient recovered. The total amount of antivenom given were 80-130 ml. Assisted ventilation were applied in cases of respiratory failure. Antibiotics covering both gram positive and negative microorganisms were also given.

3. The dermal punch biopsy were performed at the site of fang mark under aseptic technique at various hours after bite (3-96 hours). 24 out of 36 cases were done within the first 24 hours. The specimen was studied for histopathology by hematoxylin-eosin stain. Only 4 cases were studied for direct immunofluorescent test for IgG, IgM, IgA and complements.

4. The culture for both aerobic and anaerobic micro-organisms was taken immediately from the serous fluid at the biopsied sites by sterile technique in 18 cases.

Results

The histopathological sections revealed focal necrosis of the epidermis, focal collagen degeneration, lymphocytic vasculitis with fibrinoid degeneration and thrombosis of the superficial and deep

dermal vessels. There were degenerative changes of the epidermal appendages especially the eccrine sweat glands. The severity of the epidermal and dermal changes in the first 24 hours did not correlate with the time after bite. Late biopsies showed necrosis of the epidermis, dermis and subcutaneous fat. Direct immunofluorescent study showed deposition of IgM and C₃ on the wall of dermal capillaries vessels and deep dermal vessels in 2 cases. The other 2 cases were negative.

The aerobic and anaerobic bacterial cultures were taken from 18 cases (50% of the studied cases). Negative cultures were obtained in 2 cases, while most other cases (88.8%) showed mixed infection of both aerobic and anaerobic organisms. The most common organisms were gram negative bacteria.

Table 1 The aerobic bacteria grown in the studied cases.

aerobic organisms	no. of pt.	%
Proteus species	6	33.3
Providentia	3	16.6
Pseudomonas species	3	16.6
E. coli	3	16.6
Staphylococcus aureus	1	5.5
Nonhemolytic Streptococcus	1	5.5

Table 2 The anaerobic bacteria grown in the studied cases.

anaerobic organism	no. of pt.	%
Peptostreptococci	6	33.3
Clostridial species	2	11.0
Clostridial tetani	1	5.5
Propionebacterium species	1	5.5

Discussion

The histopathologic findings suggest that local reactions at the bite sites of the studied group may be caused by vasculitis and thrombosis of the superficial and deep dermal vessels. Vasculitis may be the reaction to cobra toxins, cobra antivenom or infection by micro-organisms. Since the immunologic study was done in only a few patients, it is inconclusive to suggest that the antivenom exerts its effect on the local reaction. As for the infection process, the mixed aerobic and anaerobic organisms probably played an important role

in the exertion of tissue damage. The outstanding aerobic bacteria were gram negative and very virulent. *Staphylococcus aureus* and nonhemolytic *Streptococci* were found infrequently. The anaerobic bacteria discovered were *Peptostreptococci*, *Clostridial* species and *Propriobacterium* in many cases. The proper antibiotics should cover both aerobic and anaerobic micro-organisms to prevent or treat the infection, and therefore promote faster healing. Since *Clostridial tetani* were also found, tetanus antitoxin or tetanus toxoid is indicated in snake bite patients.

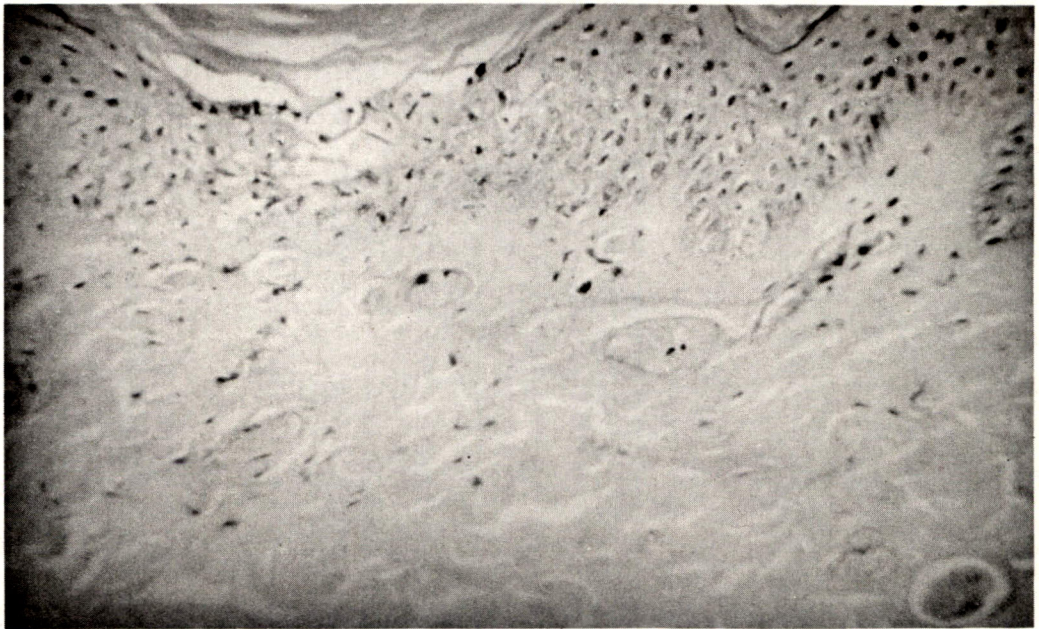


Fig. 3 Show focal necrosis of epidermis with vasculitis thrombosis and fibrinoid degeneration of superficial and deep dermal vessels (10 × 40)

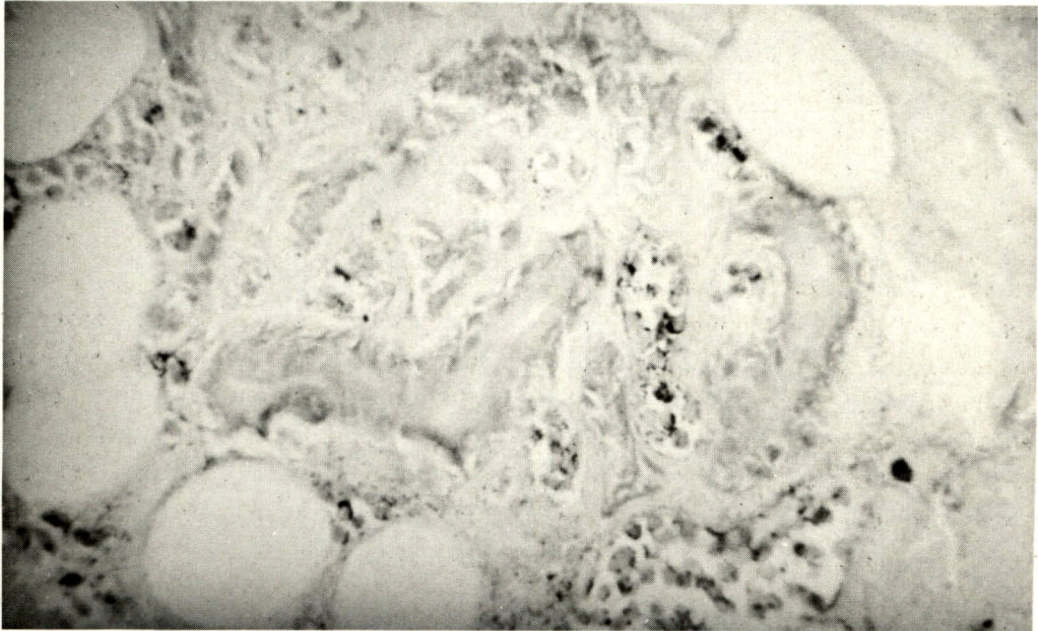


Fig. 4 Show degeneration of sweat glands (10 × 60)

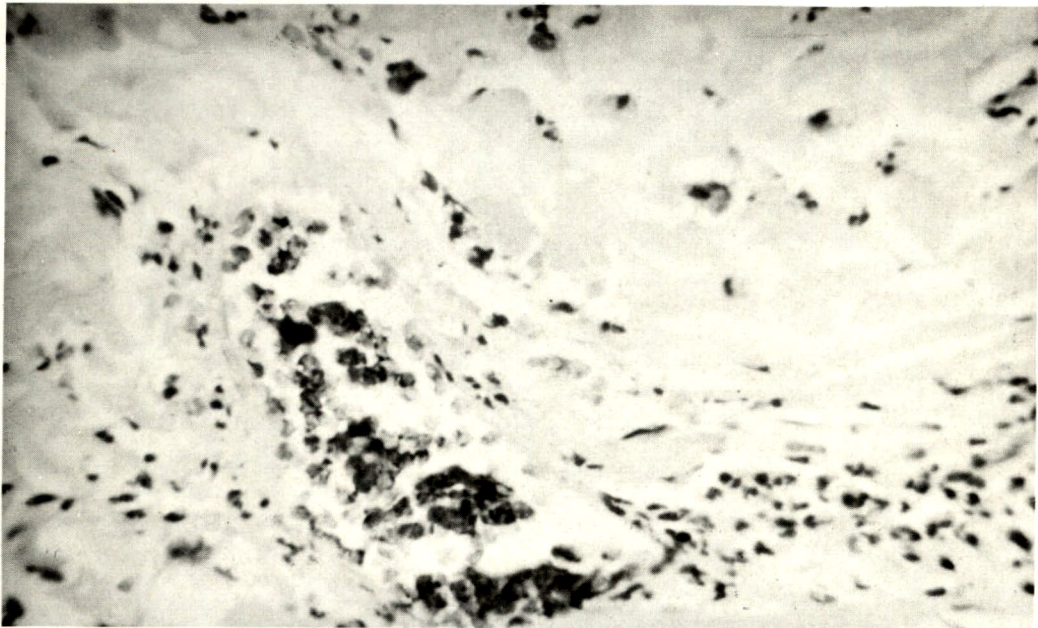
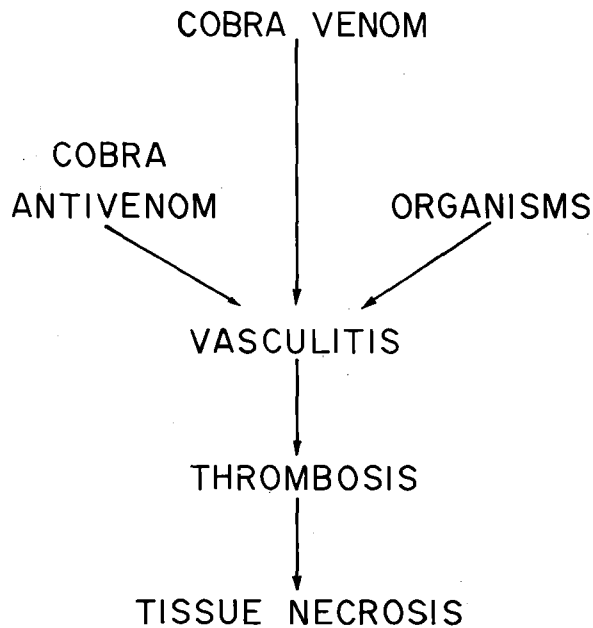


Fig. 5 Show dermal vessel with extravasation of red blood cells and leucocytoclastic vasculitis (10 × 60)



THE POSSIBLE CAUSES OF COBRA-BITE LOCAL REACTION

Summary

36 children with proven cobra bites who developed neurological manifestations were treated with cobra-antivenom intravenously. The study on histopathology, microorganism and direct immunofluorescent reaction at the sites of fang marks were carried out at different times after the bite. The histopathologic sections revealed various changes including focal necrosis of the epidermis, with degenerative change of the epidermal appendages especially the eccrine sweat glands, focal collagen degeneration, lymphocytic vasculitis, fibrinoid degeneration and thrombosis of the superficial and deep dermal vessels. In the first 24 hours the severity of the

histopathological change did correlate with the time of bite. Later biopsies certainly showed severe necrosis of the epidermis, dermis and subcutaneous fat. The bacterial cultures for both aerobic and anaerobic micro-organisms were mainly positive for *Proteus* species, anaerobic *Peptostreptococci* and *Clostridial* species. The immunologic study was done in a few cases. The histopathologic reaction may be caused by both cobra toxins and micro-organisms. Also the cobra antivenom may play some role in the local reaction.

Acknowledgements

The authors wish to thank Professor Saowani Chumdermpadetsuk for her editorial assistance.

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