

甲魚背殼前緣隆腫症組織之觀察

Histopathological Nature of the Tumorous Growth of Reared Soft Shelled Turtle, *Trionyx sinensis*

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Abstract

Epizootics of tumorous growth which caused high mortality among young reared soft shelled turtle, *Trionyx sinensis* were observed in the last several years. Light and electron microscopic study of the diseased tissues revealed that histopathological changes included: hypertrophy of the epidermal layers; destruction of the collagen fiber bundles, disintegration of chromatophores, scattering of the pigment granules, and infusion of blood corpuscles in the hypodermal layer. Degenerated blood corpuscles and mast cells were present in the hypodermal tissue. The degenerated blood corpuscles and mast cells were all showed an intact cytoplasmic membrane, pyknotic nucleus, partial or complete missing of the cytoplasm. Evidence indicated that the out growth was caused by hypertrophy associated with ulceration, not a neoplastic growth.

緒 言

甲魚 (*Trionyx sinensis* Wiegmann) 在中國自古以來即被認為是營養價值高及具多種醫療作用之補品，因為其繁殖及生長率低，甚難普及。近年來由於臺灣水產養殖業之快速發展，也提高了甲魚養殖之興趣及條件，根據余廷基在 1979 年之報導，甲魚與其他水產養殖類同樣受到許多環境因子及疾病之侵擾，嚴重者如由黴菌引起之頸腫常導至全池甲魚之死亡，造成重大之損失⁽¹⁾。農委會為了保障甲魚養殖戶之利益及提高品質，提供了對甲魚研究之條件。近年來養殖之甲魚在幼期常發生背殼前緣與頸接處產生腫瘤狀組織，一兩週內即死亡。發生之原因不明，亦未見關於此病之報導。本報告即為關於這一類似腫瘤狀組織之初步觀察之結果提供做為對於甲魚疾病有興趣者進一步研究之參考。

材 料 與 方 法

幼小甲魚背殼與頸接處發生隆腫現象者由池中撈取，放於室內之水池中，待其腫處漸大而近死亡時，將整個甲魚放置於百分之三的戊二醛固定液中 24 小時後，將病組織切下，同時於病組織近處之正常組織亦同樣取樣切成小塊 1~2 mm³，用磷酸鹽緩衝液沖洗後再放於百分之一的鐵酸中固定 1~2 小時。經過緩衝液沖洗後在乙醇中脫水，包埋於環氧樹脂中⁽²⁾，在室溫中經過 12 小時後放置於 40 度攝氏之恆溫箱中 24 小時後，轉入 60 度以上攝氏之恆溫箱中經過 72 小時後待其硬化。然後用玻璃刀在

LKB 超薄切片機上切成少於 100 nm 之薄片，用銅網撈取後在醋酸鈾 (Uranyl acetate) 及檸檬酸鉛 (lead citrate) 中⁽⁵⁾ 染色後，在日製 JEOL-10[°]CX 電子顯微鏡下觀察照相，光學顯微鏡則用石蠟包埋，甲基藍染色。

結 果

由養殖戶取得銅錢大小之幼甲魚，背殼長 3.5 cm，寬 3.0 cm，幾乎呈圓形，病患處在背殼前緣與頸部相接處，病患處為一約 4~5 mm 高，1 cm 直徑之圓形腫突之組織 (圖一)，此一組織在背殼底下，將背殼隆起但並未突破，在光學顯微鏡下，由正常與病患處皮層組織之比較 (圖二與三) 可以看出



Fig. 1. Newly hatched soft shelled turtle *Trionyx sinensis* showed tumorous growth (arrow) on the front edge of the back shell. $\times 2$



Fig. 2. Normal skin close to the tumorous growth of soft shelled turtle, it shows cuticle (C) and tightly packed epidermal layers. The major component of the hypodermal layer is collagen fiber (CF). $\times 260$

在正常組織中表皮外層之角質層甚厚，角質層以下之表皮組織，細胞層區分不明顯且緊密排列在一起，
下皮組織主要由膠原纖維束構成（圖二）。病患處皮膚之表皮組織中角質層消失。表皮細胞層中的細胞
略成肥大，細胞區分明顯，下皮層中的膠原纖維減少或消失，組織有破裂處（圖三）。

以上之變化在較高倍放大影像下差異更為明顯（圖四），在正常狀態下皮膚上之皺折伏在皮膚之表

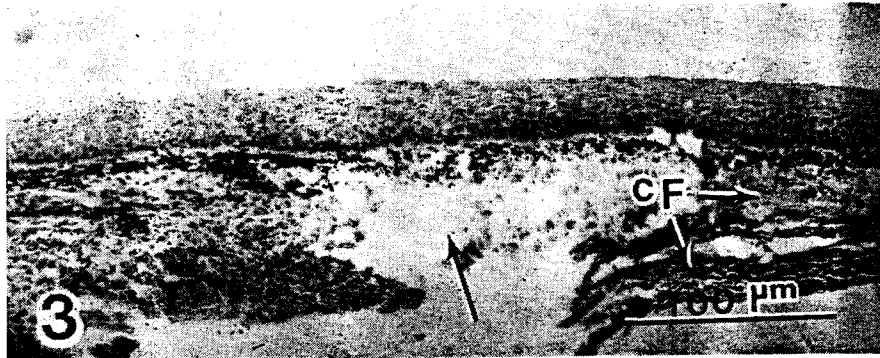


Fig. 3. Skin from the area of tumorous growth shows reduced cuticle, hypertrophy epidermal layers, the amount of collagen fiber (CF) is greatly reduced, arrow indicated a broken spot of the hypodermal layer. $\times 260$

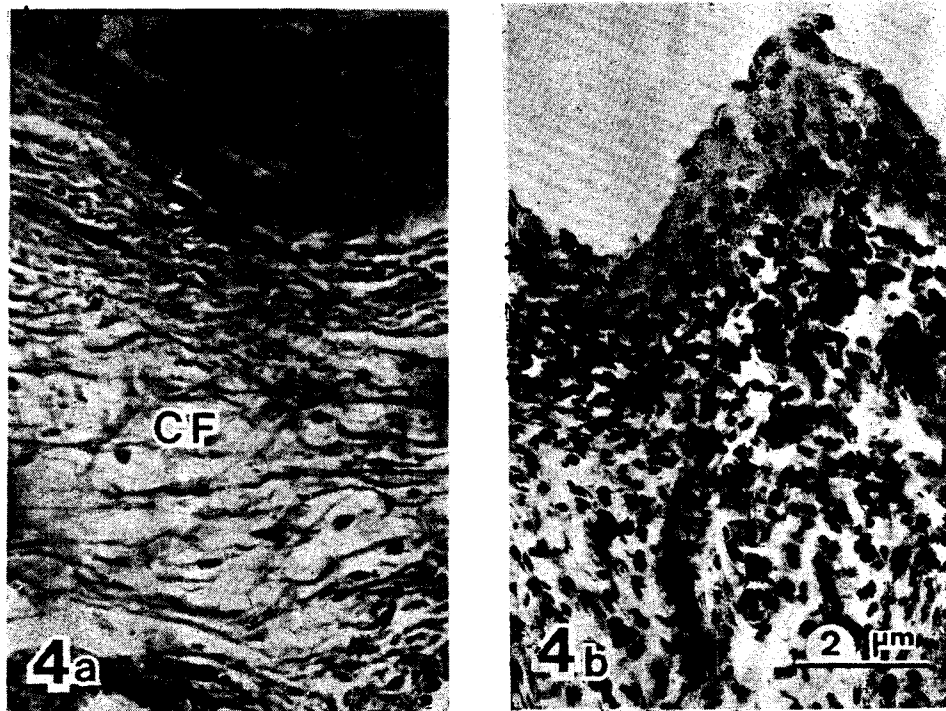


Fig. 4. Higher magnification of the skin of both normal (a) and with tumorous growth (b). The epidermal layer of the normal skin shows a fold of the skin. Similar fold in (b) is left up due to hypertrophy of the epidermal cells. The collagen fibers (CF) in (a) of the hypodermal layer is missing in (b). The hypodermal layer in (b) is replaced by blood cell infusion. $\times 800$

皮(圖四 a)，但在病患部位之皺折亦因細胞之肥大而直立(圖四 b)，下皮層在病患部位膠原纖維之消失亦至為明顯。

在電子顯微鏡下，下皮組織在正常的情况下膠原纖維束結構緊密，色素體^(2,3)含在色素細胞中，色素細胞之伸延部份位於膠原纖維束之間(圖 5a)，病患部位相似位置之下皮組織膠原纖維束破損，結構不完整，色素細胞之伸延部分亦已破損，色素體散佈於膠原纖維束之間的空隙中(圖 5b)。

在破損的皮層中充滿了退化之血球細胞，紅血球細胞多為由細胞膜包圍着的濃縮細胞核，細胞核呈方形細胞質已完全消失(圖 6, 7a 及 7b)，白血球多尚殘留部分細胞質在內，細胞膜完整，細胞核位於細胞之中央，異染色質位於細胞核膜之邊緣上(圖 6, 7a)。此外尚可觀察到巨細胞，退化了的巨細胞僅為細胞膜包圍着的顆粒體及細胞核，顆粒體排列在細胞核之周圍，核與膜之間的空隙，細胞質已不存在，細胞核中的異染色質在於核膜之邊緣(圖 7b)。在皮層之組織中並無新生腫瘤組織之存在，皮膚之隆起為表皮細胞之肥大所致。

討 論

甲魚背殼隆腫的起因不明。在觀察過程中雖無做病原的分離實驗，但亦無觀察到類似細菌、黴菌或病毒體的存在，正如結果中提到，此一隆腫的現象並非一衍生的腫瘤組織而是由於細胞的肥大與充血而成。雖然在此刻無法確定致死的因子為此一隆腫的組織，但在甲魚垂死之時，此一隆腫組織為唯一觀察

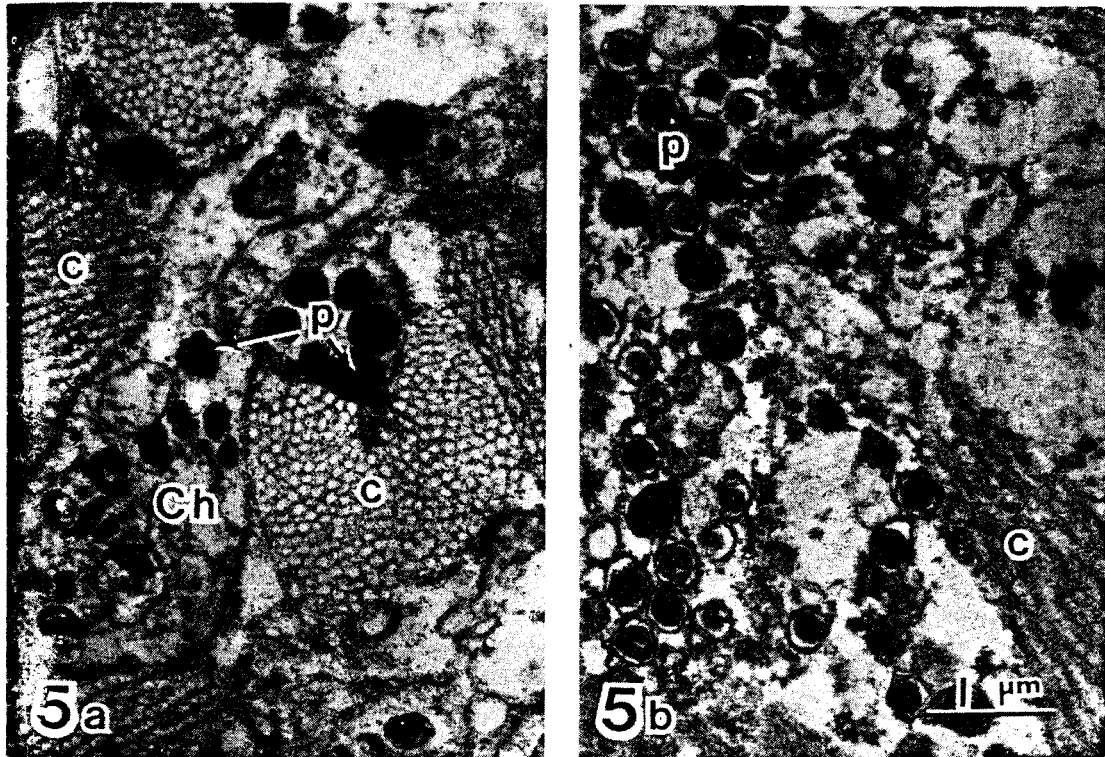


Fig. 5. Electronmicrographs of the hypodermal layers from both normal (a) and (b) diseased skin of soft shelled turtle. In (a) the collagen fiber bundles (C) are well organized, pigments (P) in the chromatophores (Ch) between the collagen bundles. In (b) the collagen fiber bundles are disorganized (C), and the pigments (P) are scattered between the disorganized collagen bundles. $\times 15,000$

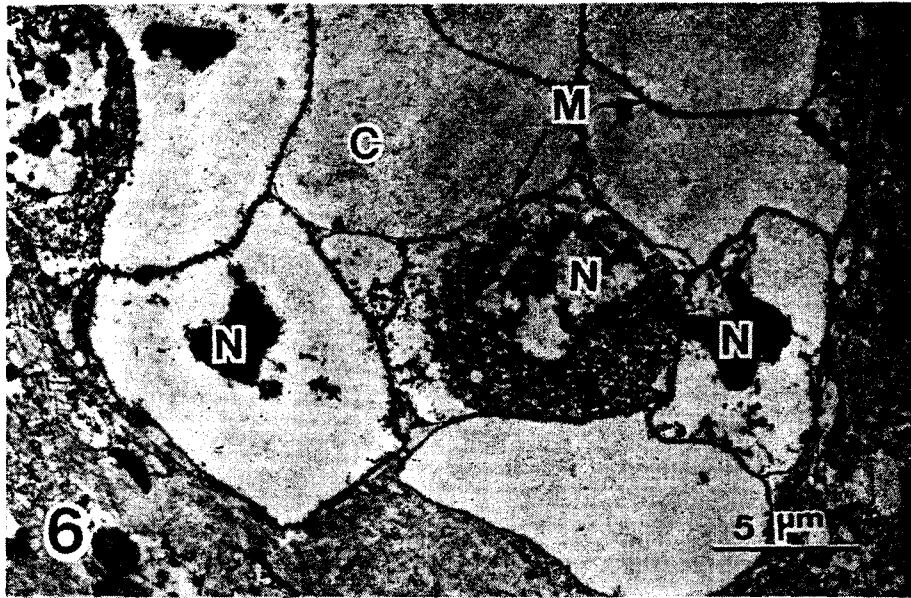


Fig. 6. Electronmicrograph of the hypodermal layer from area of tumorous growth shows a broken spot of the hypodermal layer filled with degenerated blood cells. The degenerated red blood cells each shows a pyknotic nucleus (N), empty cytoplasmic space (C), surrounded by an intact cytoplasmic membrane (M). The white blood cell with a central located nucleus with marginated heterochromatin and granulated cytoplasm. $\times 3,500$

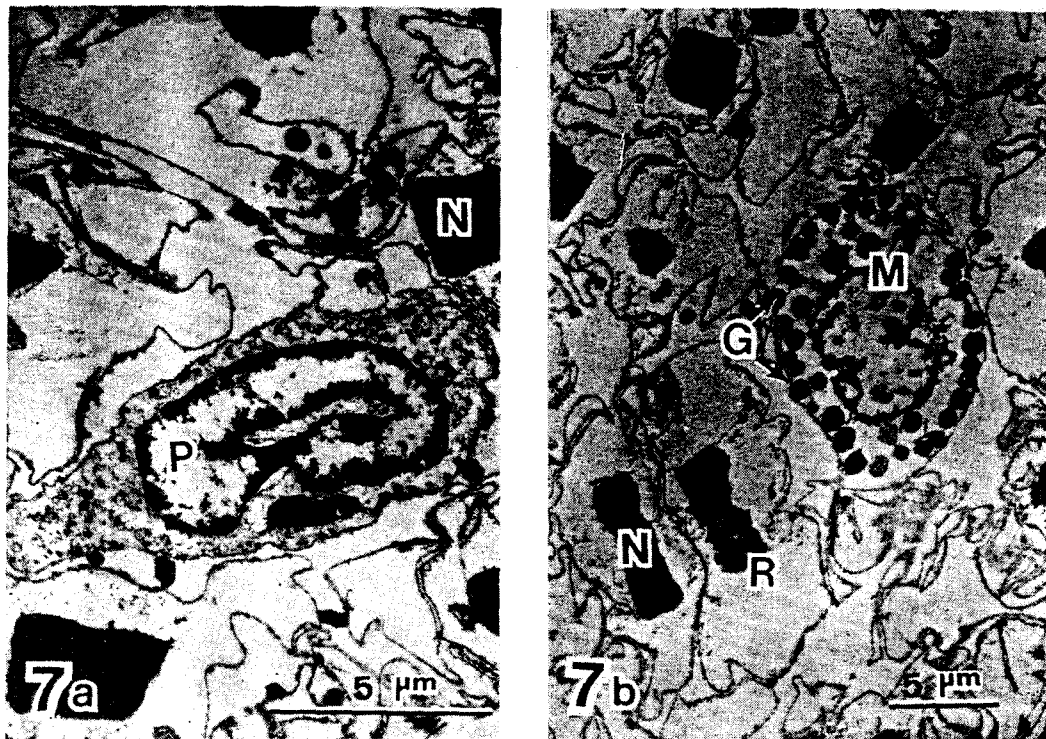


Fig. 7. Electronmicrographs of the degenerated blood cells within the broken spots of the hypodermal layer of the diseased area. Among the degenerated red blood cells (R) which show central located, pyknotic rectangular shaped nucleus (N) there are polymorphonuclear leukocyte (P) (a) and degenerated mast cell (M) with a central located nucleus with marginated heterochromatin and granules (G) (b). All these cells are still surrounded with an intact cell membrane. a. $\times 5,600$ b. $\times 2,800$

到的病患，且內部呈潰瘍性病變。本報告僅完成此一病患處皮層之組織情況與病患組織引起之變化，提供有對於此一病患有興趣者做進一步之研究。

摘 要

養殖甲魚 (*Trionyx sinensis*) 在發育期，發生大量死亡，死亡時的病徵為甲殼前緣與頸臨接處，發生腫起潰瘍的現象。利用光學與電子顯微鏡對病症處與正常之組織做比較性的觀察。

觀察的結果發現，此一地區的細胞均成隆大現象，血球在此一部位壞死現象變化中，亦經過衰退而有特殊的構造，色素細胞及膠原纖維在此組織中之排列情形，均作了初步的討論。

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