Case Report

Cisplatin Intralesional Chemotherapy of Recurrence Schwannoma in Horse: A Case Report

Rungrueang Yodsheewan[1] Kanittha Phetudomsinsuk[2*]

Abstract

A 7-year-old crossbred stallion had multiple solitary cutaneous masses at the right infraorbital area which were recurrent for two months. The biggest mass size was 5 x 7 x 1 cm^3 and had superficial bleeding. This case was diagnosed as peripheral nerve sheath tumors based on cytological and histological examination which showed numerous spindle cells with cancerous characteristics, cell patterns of the tumor were interlacing bundle and a whirl. Most areas showed tense palisading nuclei of tumor cells (Verocay bodies) known as Antoni A area. The rest showed loose paucicellular tissue known as Antoni B area. Within histologic sections, the cells showed positive immunoreactivity for S100 and calretinin, whereas immunoreactivity was not detected for CD34. Rapidly recurrent tumor was found after excision combined with cryotherapy. Consequent treatment using cisplatin (1mg/cm^3) in autologous serum was intralesionally injected twice, 2 weeks apart. After the two injections, the horse responded with effective wound healing two months later. Therefore, intra-lesional injection of cisplatin should be considered for recurrent dermal Schwannoma or inadequate surgical removal of tumor.

Keywords: cisplatin, equine, Schwannoma

[1]*Department of Pathology, Faculty of Veterinary Medicine, Kasetsart University, Kamphaeng Saen Campus, Nakhon Pathom, Thailand, 73140*

[2]*Department of Large Animal and Wildlife Clinical Sciences, Faculty of Veterinary Medicine, Kasetsart University, Kamphaeng Saen Campus, Nakhon Pathom, Thailand, 73140*

***Correspondence**: fvetktp@ku.ac.th*

Introduction

Equine cutaneous masses are classified as neoplastic masses (sarcoids, squamous cell carcinoma, fibroma, and melanoma) and non-neoplastic masses (Carstanjen et al., 1997). Sarcoid and squamous cell carcinoma occur more frequently in equine neoplasm but cutaneous benign peripheral nerve sheet tumors (PNSTs) in dogs, cats, horses, and cattle are rare (Goldschmidt and Hendrick, 2002; Valentine, 2006; Schöniger and Summers, 2009).

Schwannomas and neurofibromas are the most common benign peripheral nerve neoplasms. Schwannomas are relatively uncommon slow-growing tumors that originate from the Schwann cells of the nerve roots, while neurofibromas display a mixture of cell types, including Schwann cells, perineurial cells, and endoneurial fibroblasts. The etiology of Schwannoma is still unknown and is generally asymptomatic (Enoz et al., 2006). Gross characteristics of Schwannoma are firm to soft, well-circumscribed, unencapsulated masses in the dermis as single or multiple masses. Schwannomas in horses may occur at the age of 3 to 16 years, no breed or sex predisposition (Fernandez et al., 1996). In domestic animals, benign PNSTs occur more commonly on the eyelids (Pascoe and Summers, 1981; Schöniger et al., 2011) and, in humans, mostly occur in the head and neck region (Pandarakalam et al., 2005).

Histopathology of Schwannoma is a whirl of spindle-shaped to ovoid with elongated cytoplasmic extensions in a collagenous matrix form Antoni A and Antoni B areas, which could be differentiated from equine sarcoid (Goldschmidt and Hendrick, 2002). In general, Schwannoma may be misdiagnosed as fibroma, fibrosarcoma or sarcoid (Scott and Miller, 2003).

Histological characteristic features with immunohistochemistry for cellular markers are usually diagnosed in human Schwannoma and neurofibroma (Weiss and Goldblum, 2008; Skovronsky and Oberholtzer, 2004). Schwann cells were immunopositive for S100 protein and expression of laminin and glial fibrillary acidic protein (Schöniger et al., 2011).

Surgical resection is the principal treatment for skin tumors; however cutaneous tumors in horses have poorly defined margins, possibly local infiltrations and high recurrence rates. Various combining treatments include the use of a Nd:YAG or CO_2 laser (McCauley et al., 2002); intralesional chemotherapy with cisplatin (Hewes and Sullins, 2006) or 5-fluorouracil; cryotherapy; and brachytherapy (Saulez et al., 2009). Using Nd:YAG laser and brachytherapy gives highly successful results in periorbital neoplasms. However, the high cost of instruments, the need for professional technician and the hazard from radioisotope are the restrictions (Byam-Cook et al., 2006). Although cryotherapy is an easier and cheaper method compared to Nd:YAG laser and brachytherapy, the recurrence of tumor is in a higher ratio. Cisplatin oil emulsion was developed for equine skin tumor therapy (Theon et al., 1993), combined with electrical treatment it becomes highly potent (Tamzali et al., 2012).

The purpose of this study was to report the success of injections of cisplatin intralesional chemotherapy without sesame seed oil emulsion.

Case History

A 7-year-old crossbred stallion was presented to the Kasetsart University Veterinary Teaching Hospital, Kamphaeng Saen Campus, with multiple nodular dermal masses that became larger within 2 months after an initial excision by the owner. The stallion was bright, alert and in good condition at the time of presentation. Normal cardiovascular and respiratory parameters were shown. The visual reflex, appearance and signs were always normal. There were externally visible, multiple fleshy nodules at the right infraorbital. Palpation of the right infraorbital revealed a subcutaneous swelling. The biggest expansive nodular mass was 5 x 7 x 1 cm^3 with multifocal small hemorrhage (Fig 1A). Squamous cell carcinoma, fibrosarcoma, and sarcoid were diagnosed based on the clinical signs and gross lesion.

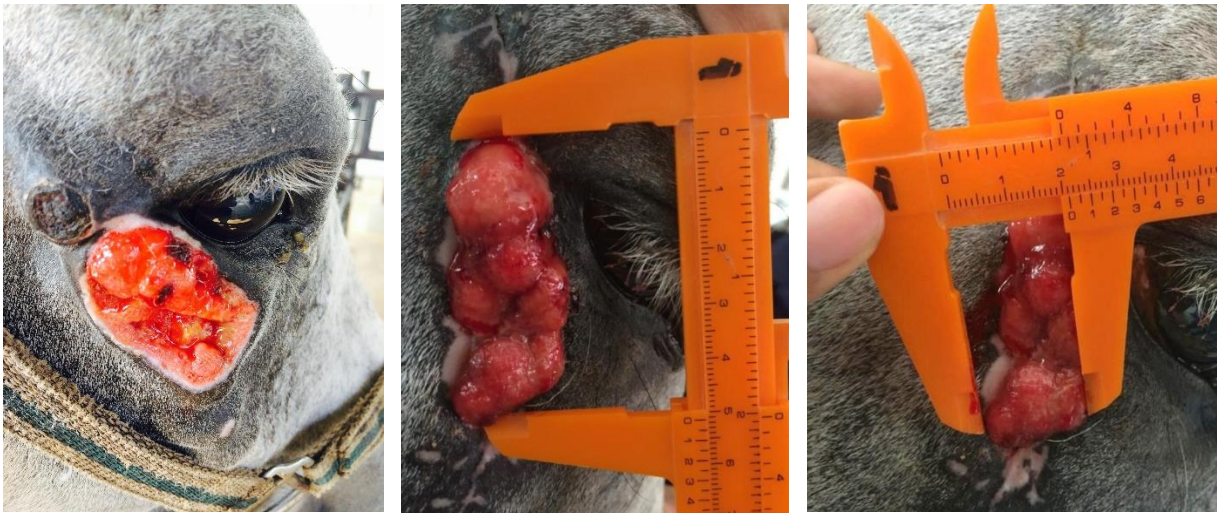

Figure 1 Gross lesion finding demonstrated nodules at the right infraorbital. **A**, on the day presented. **B-C**, at day 2 after excision.

Diagnostic Method

Fresh surgical specimens of an infraorbital mass were fixed in 10% w/v buffered formalin, embedded in paraffin and stained with Hematoxylin and Eosin (HE). Histological section was prepared and stained for histological feature at Kasetsart University.

Fine needle aspiration, stained with Modified Wright's staining, revealed a great number of spindle-shaped cells characterized by dense chromatin, 1-3 prominent nucleoli, anisokaryosis, anisocytosis, and basophilic cytoplasm. In addition, many eosinophils and other inflammatory cells were observed (Fig 2). As a result, cytological investigation suggested mesenchymal cell tumor. The histopathological findings demonstrated hemorrhagic ulcerative dermatitis due to rupture of epidermis, hemorrhage, and infiltration of inflammatory cells which were mainly eosinophils and neutrophils. The mass was non-encapsulated, poorly demarcated, and was composed of slightly pleomorphic fusiform cells with oval-shaped nuclei containing intracytoplasmic vacuoles in some cells. The cells were arranged in interlacing bundle and whirling pattern. Most areas showed abundant palisading nuclei of tumor cells known as Antoni A area. The rest showed loose paucicellular tissues known as Antoni B area (Fig 3). Histopathological diagnosis was benign peripheral nerve sheath tumor.

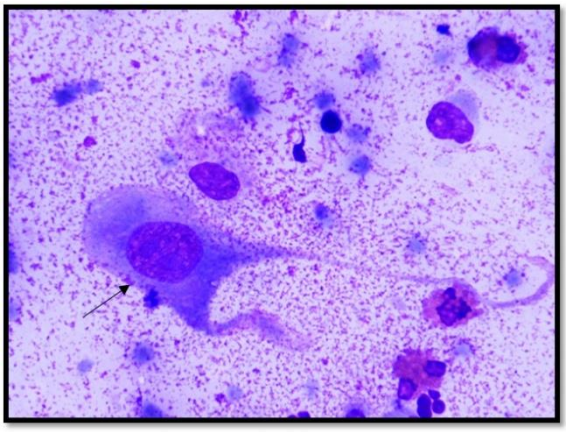

Figure 2 Cytology revealed spindle-shaped cells characterized by dense chromatin, 1-3 prominent nucleoli, anisokaryosis, anisocytosis, and basophilic cytoplasm (black arrow).

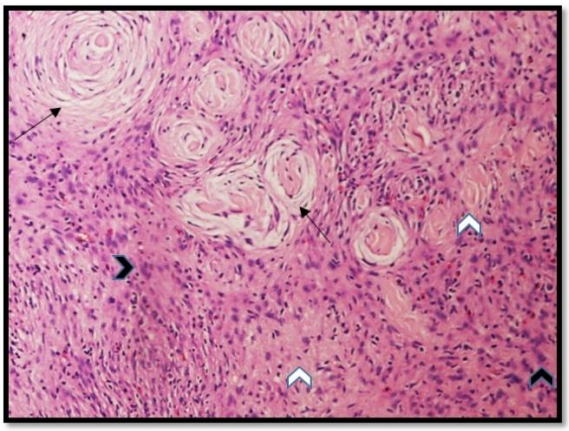

Figure 3 Histopathological findings demonstrated that the cells were arranged in interlacing bundle and whirling pattern (black arrow). Most areas showed abundant palisading nuclei of tumor cells known as Antoni A area (black arrow head). The rest showed loose paucicellular tissues known as Antoni B area (white arrow head).

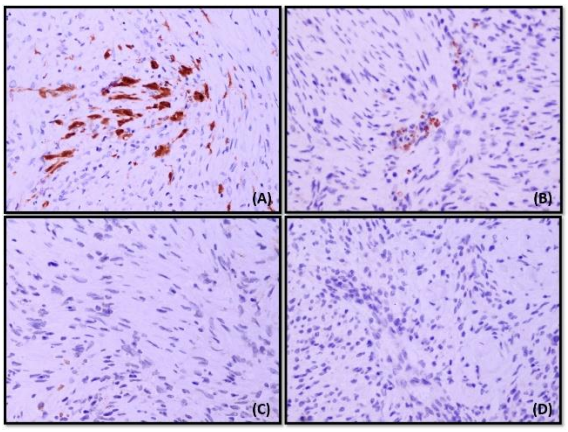

Figure 4 Immunohistochemistry of the mass. **A**, S100 protein was positive as intense cytoplasmic staining. **B**, calretinin staining was positive as brown spot. **C**, CD34 marker staining was negative and **D**, negative control.

Immunohistochemistry

The diagnosis of Schwannoma was confirmed using antibodies against S100 protein (anti-S100 mouse monoclonal antibody, Cell Marque, USA), calretinin (anti-calretinin rabbit monoclonal antibody, Cell Marque, USA), and CD34 (anti-C34 mouse monoclonal antibody, Neo MARKER, USA) from Institute of Pathology, Thailand. For S100 and CD34, indirect immunohistochemistry method was performed with 1:100 dilution of primary antibodies and detected by peroxidase enzyme system, while for calretinin labeled streptavidin-biotin (LASB) method was used with 1:100 dilution of primary antibody and detected by peroxidase enzyme system. A characteristic histologic feature with immunohistochemistry for S100 was strongly positive, calretinin was positive and CD34 was negative (Figs 4A-4D). The positive results for S100 protein and calretinin suggested Schwannoma.

Treatment

The initial treatment was surgical removal and suture. The wound was dehiscence and hypergranulation tissue was present. Excision combined with topical cryotherapy, metal probe cooled with liquid nitrogen directly touching the skin over a period of 2 min, was performed two weeks later. This treatment was ineffective again; there were recurrent multiple nodules and continuous growth within 48 h after excision. The size was 2 x 5 x 1 cm^3 (Figs 1B-C). The horse was restrained and premedicated with acepromazine (0.04 mg/kg, iv.) and xylazine hydrochloride (0.5 mg/kg, iv.) and auriculopalpebral and supraorbital nerves were blocked with 2% lidocaine. Intralesional administration of cisplatin 1 mg/cm^3 in autologous serum (2:1 ratio) was injected into the lesion twice at two-week intervals. The injection was spaced 6 mm apart in 2 planes and in a 1 cm wide outwardly healthy margin.

Results and Discussion

Only surgical removal or excision combined with cryotherapy was unsuccessful; the mass recurred within 48 h. Intralesional cisplatin was chosen for additional therapy. The intralesional cisplatin injection twice at 2-week intervals was performed. There were no oedematous reaction located on thin skin regions, healing delay and recurrence in this stallion after two months. However, the mass should be followed up at a minimum of 6 months.

Histopathology findings was a whirl of spindle to ovoid shape with elongated cytoplasmic extensions in a collagenous matrix form Antoni A and Antoni B areas which are the characteristics of benign peripheral nerve sheath tumors (PNSTs). In veterinary literature, PNSTs, Schwannoma and neurofibroma were not clearly classified (Koestner et al., 1999; Scott and Miller, 2003). All of these terms were used interchangeably. However, distinction of the tumors is very important for prognosis and treatment.

Immunohistochemistry for S100 protein markers have been studied for separating neural tumors from nonneural origin tumors and also for differentiating Schwannomas from neurofibromas (Valentine, 2006; Weiss and Goldblum, 2008; Skovronsky and Oberholtzer, 2004). Calretinin is a 29-kd, calcium-binding protein expressed primarily in reliable types of neurons in the central and peripheral nervous systems. Samson et al. (2004) demonstrated that calretinin staining was positive in 24 of 25 (96%) Schwannomas and, therefore, this staining was useful for differentiating Schwannomas from neurofibromas. CD34 is a transmembrane glycoprotein normally expressed by lymphoid and myeloid lineage (Civin et al., 1984). Anti-CD34 antibodies also react with endothelial cell, periadnexal spindle cell and dendritic cells (Nickoloff, 1991). In human, anti-CD34 antibody was used in the immunohistochemistry method to exclude tumor from Schwann cell origin from other PNSTs because it can react with dendritic cell within endoneurium of peripheral nerve but not react with conventional Schwannoma (Weiss and Nickoloff, 1993).

To our knowledge, the CD34 markers are usually used for differentiating human Schwannomas and there are no reports of this marker in horse. The positivity for both S100 protein and calretinin as well as the negative immunolabeling for CD34 marker in this case could suggest that the tumor was from Schwann cell origin.

There are several treatments for benign cutaneous tumor (Carstanjen et al., 1997). Surgical resection is a primary treatment, however cutaneous tumors in horses tend to have poorly defined margins, may be locally infiltrative and have a high recurrence rate. Fifty per cent of periocular Schwannomas recurred following surgical removal within a 6-month period (Saulez et al., 2009), similar to this case. Additional therapies including laser, photodynamic therapy, radiation therapy, brachytherapy, and chemotherapy should produce more satisfied results. Saulez et al. (2009) reported the success of interstitial brachytherapy using Ir192 for malignant buccal Schwannomas. Cisplatin intralesional injection is a successful treatment for human Schwannoma (Zhang et al., 2013). However, cisplatin was 83% (40/48) successful in treating cutaneous neoplasia in horses, which did not recur in 2 years (Hewes and Sullins, 2006) and 99.5% (193/194) in treating sarcoid tumor, which did not recur in 4 years (Tamzali et al., 2012). Cisplatin solution should be prepared immediately prior to use with mixture of cisplatin, medical grade of sesame oil and autologous serum (Theon et al., 1993). Sequential therapeutic of cisplatin oil emulsion on large tumor was reported by Hews and Sullins (2006) and Theon et al. (2007). This study showed that intra-lesional cisplatin without sesame oil after 2 treatments provided satisfied results. The suspension without sesame oil decreasing the frequency of treatment might be due to the increase in intracellular concentrations.

References

Byam-Cook KL, Henson FM, Slater JD. 2006. Treatment of periocular and non-ocular sarcoids in 18 horses by interstitial brachytherapy with iridium-192. Vet Rec. 159(11): 337-41.

Carstanjen B, Jordan P, and Lepage OM. 1997.Carbon dioxide laser as a surgical instrument for sarcoid therapy--a retrospective study on 60 cases. Can Vet J. 38(12): 773–776.

Civin CI, Strauss LC, Brovall C, Fackler MJ, Schwartz JF and Shaper JH. 1984. Antigenic analysis of hematopoiesis, III: a hematopoietic progenitor cell surface antigen defined by a monoclonal antibody raised against KG-1a cells. J Immunol. 133: 157-165.

Enoz M, Suoglu Y and Ilhan R. 2006. Lingual Schwannoma. J Cancer Res Ther. 2(2): 76–78.

Fernandez CJ, Valentine BA, Smith C and Summers BA. 1996. Equine dermal Schwannoma. Veterinary Pathology 33: 607.

Goldschmidt MH and Hendrick MJ. 2002. Tumors of the skin and soft tissues. In: Tumors in Domestic Animals, ed. Meuten DJ, 4th ed. Ames, IA: Iowa State Press. 45–117.

Hewes CA and Sullins KE. 2006. Use of cisplatin-containing biodegradable beads for treatment of cutaneous neoplasia in equidae: 59 cases (2000–2004). J Am Vet Med Assoc. 229(10): 1617-1622.

Koestner A, Bilzer T, Fatzer R, Schulman FY, Summers BA and Van Winkle TJ. 1999. Tumors of the peripheral nervous system. In: Histological Classification of Tumors of the Nervous System in Domestic Animals. 2nd (series) vol. 5. Washington DC: Armed Forces Institute of Pathology. 36–38.

McCauley CT, Hawkins JF, Adams SB and Fessler JF. 2002. Use of a carbon dioxide laser for surgical management of cutaneous masses in horses: 32 cases (1993-2000). J AmVet Med Assoc. 220(8): 1192-7.

Nickoloff BJ. 1991. The human progenitor cell antigen (CD34) is localized on endothelial cells, dermal dendritic cells, and perifollicular cells in formalin-fixed normal skin, and on proliferating endothelial cells and stromal spindle-shaped cells in Kaposi's sarcoma. Arch Dermatol. 127: 523-529.

Pandarakalam C, Sudha S, Shameena PM and Varghese VI. 2005. An unusual presentation of a case of Schwannoma, J Oral Maxillofac Pathol. 9(1): 27–29.

Pascoe RR and Summers PM. 1981. Clinical survey of tumours and tumour-like lesions in horses in south east Queensland. Equine Vet J. 13 (4), 235–239.

Samson WF, Steve AMc, and Maomi L. 2004. Immunohistochemical Staining for Calretinin Is Useful for Differentiating Schwannomas From Neurofibromas Am J Clin Pathol. 122: 552-559.

Saulez, MN, Voigt A, Steyl JCA, Wilpe E, Kotzen J and Daniels F. 2009. Use of Ir192 interstitial brachytherapy for an equine malignant dermal Schwannoma. J S Afr Vet Assoc. 80(4): 264–269.

Schöniger S and Summers BA. 2009. Localized, plexiform, diffuse and other variants of neurofibroma in twelve dogs, two horses and a chicken. Vet Pathol. 46: 904–915.

Schöniger S, Valentine BA, Fernandez CJ, and Summers BA. 2011. Cutaneous Schwannomas in 22 Horses. Vet Pathol. 48(2): 433-442.

Scott DW and Miller WH Jr. 2003. Neoplastic and non-neoplastic tumours. In Equine dermatology. St Louis: W B Saunders. 698–795.

Skovronsky D and Oberholtzer JC. 2004. Pathologic classification of peripheral nerve tumors. Neurosurg Clin N Am. 15:157–166.

Tamzali Y, Borde L, Rols MP, Golzio M, Lyazrhi F and Teissie J. 2012. Successful treatment of equine sarcoids with cisplatin electrochemotherapy: a retrospective study of 48 cases. Equine Vet J. 44(2): 214-220.

Theon AP, Pascoe JR, Carlson GP and Krag DN. 1993. Intratumoral chemotherapy with cisplatin in oily emulsion in horses. J Am Vet Med Assoc. 202: 261–267.

Theon AP, Wilson WD, Magdesian KG., Pusterla N, Snyder JR and Galuppo LD. 2007. Long-term outcome associated with intratumoral chemotherapy with cisplatin for cutaneous tumours in equidae: 573 cases (1995-2004). J Am vet med Assoc. 230: 1506-1513.

Valentine BA. 2006. Neoplasia. In: Equine geriatric medicine and surgery Bertone J (ed). St Louis: W B Saunders. 147–167.

Weiss SW and Goldblum JR. 2008. Benign tumors of peripheral nerves. In: Enzinger and Weiss's Soft Tissue Tumors, 5th ed. JR Goldblum, SW Weiss and AL Folpe (ed). St Louis MO: Mosby Elsevier. 825–901.

Weiss SW and Nickoloff BJ. 1993. CD-34 is expressed by a distinctive cell population in peripheral nerve, nerve sheath tumors, and related lesions. Am J Surg Pathol.;17: 1039-1045.

Zhang SQ, Wu S, Yao K, Dong P, Li YH, Zhang ZL, Li XX and Zhou FJ. 2013. Retroperitoneal Schwannoma mimicking metastatic seminoma: case report and literature review. Chin J Cancer. 32(3): 149-152.

บทคัดย่อ

การรักษาเนื้องอกชวานโนมาแบบเกิดซ้ำในม้าโดยใช้เคมีบำบัดด้วยยาซิสปาติน: รายงานสัตว์ป่วย

รุ่งเรือง ยอดชีวัน[1] ขนิษฐา เพชรอุดมสินสุข[2*]

พ่อม้าพันธุ์ผสมอายุ 7 ปีมีก้อนเนื้อใต้ผิวหนังเกิดซ้ำที่บริเวณใต้เบ้าตาขวามาเป็นระยะเวลา 2 เดือน ก้อนเนื้อมีขนาด 5x7x1 ลูกบาศก์เซนติเมตรและผิวหน้ามีเลือดออก สัตวแพทย์ทำการตัดก้อนชิ้นเนื้อเพื่อศึกษาลักษณะทางเซลล์และจุลพยาธิวิทยา ผลการตรวจทำให้สงสัยว่าเป็นเนื้องอกของกลุ่มเซลล์ในระบบประสาทเนื่องจากพบเซลล์รูปทรงกระสวยจำนวนมากซึ่งมีลักษณะของเซลล์มะเร็ง โดยเซลล์มะเร็งจัดเรียงตัวเป็นแถวสานต่อเนื่องกันเป็นมัดๆ และบางบริเวณหมุนวนเป็นก้นหอย มีสองบริเวณที่เห็นได้ชัดเจนคือ บริเวณที่มีเซลล์มากนิวเคลียสเรียงตัวกันเป็นแถวหรือเวโรเคย์บอดี เรียกพื้นที่บริเวณนี้ว่าแอนโทไนท์เอ และบริเวณที่มีเซลล์น้อยเรียกแอนโทไนท์บี จึงทำการตรวจเพิ่มเติมโดยการใช้แอนติบอดีและพบว่าเซลล์เนื้องอกให้ผลบวกต่อโปรตีนและคาลเรตินิน แต่ให้ผลลบกับแอนติบอดีต่อซีดีสามสิบสี่ จากการวินิจฉัยจึงสรุปได้ว่าม้าเป็นเนื้องอกชนิดชวานโนมา การรักษาในขั้นต้นได้ทำการตัดชิ้นเนื้องอกออกร่วมกับการทำไครโอเธอราปี ภายหลังการรักษาเพียงสองวันพบว่าเนื้องอกเจริญขึ้นซ้ำที่บริเวณวิการเดิม สัตวแพทย์จึงทำการรักษาด้วยการตัดเนื้องอกร่วมกับการให้ยาซิสปาตินผสมซีรั่มฉีดเข้าตำแหน่งวิการ 2 ครั้ง ห่างกันครั้งละ 2 สัปดาห์ ภายหลังการฉีดด้วยซิสปาตินเข็มที่หนึ่งพบว่าวิการของแผลแห้ง และหลังการฉีดซิสปาตินเข็มที่สอง 2 เดือนไม่พบการเจริญขึ้นซ้ำของเนื้องอก ดังนั้นการฉีดซิสปาตินเข้าตำแหน่งวิการสามารถรักษาเนื้องอกชวานโนมาที่มีอุบัติการเกิดซ้ำหรือไม่สามารถรักษาได้ด้วยวิธีการตัดออกเพียงอย่างเดียว

คำสำคัญ: ซิสปาติน ม้า เนื้องอกชวานโนมา

[1]ภาควิชาพยาธิวิทยา คณะสัตวแพทยศาสตร์ มหาวิทยาลัยเกษตรศาสตร์ วิทยาเขตกำแพงแสน จ.นครปฐม 73140

[2]ภาควิชาเวชศาสตร์คลินิกสัตว์ใหญ่และสัตว์ป่า คณะสัตวแพทยศาสตร์ มหาวิทยาลัยเกษตรศาสตร์ วิทยาเขตกำแพงแสน จ.นครปฐม 73140

****ผู้รับผิดชอบบทความ** E-mail: fvetktp@ku.ac.th*