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DERMATOPHYTOSIS IN CATTLE: CLINICAL, HISTOPATHOLOGICAL AND THERAPEUTIC STUDIES

(With 4 Tables and 6 Figures)

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دراسات اكلينيكية وياتولوجية وعلاجية عن القراع في الأبقار

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تم إجراء هذه الدراسة التعرف على بعض الجوانب الوبائية والمناعية والتشخيصية والعلاجية لمرض القراع في الأبقار، بالفحص المعملي ثم الكثيف عن فطر التريكوفيتون بنسبة لمرض القراع في الحيوانات التي تم فحصها. كانت الإصابة عبارة عن دوائر خالية من الشعر في مناطق مختلفة من الجلد خاصة في مناطقة الرأس وحول العينين ومنطقة الأنن والرقبة. كانت نسبة الإصابة عالية في فصلي الثناء والربيع خاصة في العجول الصغيرة في المرحلة العمرية ما بين ٣ أشهر وعام. اظهرت النتائج المعملية أيضا وجود ارتفاع معنوي في مستوى الجما جلوبيولين وكذلك مستوى الأجسام المصادة IgG في الحيوانات السليمة عنها في الحيوانات المسليمة عنها في الحيوانات المحابة، تم أيضا تقييم كفاءة عسول يستخدم في العلاج بمقارنته بمحلول صبغة اليود المركز ٥٠٠ حيث أعطى الغسول كفاءة عالية في العلاج.

SUMMARY

Out of 486 cattle examined for ringworm, 107(22.01%) cases proved to be infected with Trichophyton spp. by direct microscopy and culture methods. Histopahtological studies of skin biopsies before treatment revealed marked hyperkeratosis, parakeratosis, acanthosis, acantholysis and leukocytic infiltration. Higher incidence of the disease was recorded

only rarely useful (Stervenson and Hughes, 1988; Scott et al., 1995; Carlotti and Bensignor, 1999 and Markus et al., 2001).

In Egypt, dermatophytosis was studied by many authors (Abu-Gabal et al., 1976; Abdel-Halim et al., 1988; Rafaa, 1992; Seddik et al., 1994; Awad, 1995; Abou-Zaid, 1996 and Ammar et al., 1998) with incidence varied from 10.42% to 98.3%. The aim of the present work was directed to study some epidemiological, clinicl and histopathological aspects as well as sero-biochemical responses associated with dermatophytosis in cattle. Also the efficacy of Tr. Iodine and other anti fungal preparation was studied.

MATERIALS and METHODS

1) Animals:

For estimating the yearly incidence of dermatophytosis in Sakha farm during 2001, 486 animals of different ages were monitored for the occurrence of the disease (longitudinal study).

2) Mycological examination:

a- Skin scraping:

It was done according to Kely (1984).

b- Direct microscopic examination:

The scraping and hair were placed in a 20% potassium hydroxide solution on a clean slide, then covered with a cover slip and gently warmed. After appropriate amount of time for clearing the debris, the slides were examined under microscope with reduced lighting for the presence of hyphae and arthrospores as described by Koneman and Roberts (1985).

c- Culture:

Skin scraping and hair were inoculated on Sabouraud Dextrose Agar media with chloramphenicol and cyclohedidine (actidione) and Kept at 27°C for at least 3 weeks. The resultant colonies were identified according to (St-Germain and Summerbell, 1996).

3) Histopathological examination:

Skin biopsies of entire skin were taken from the affected parts as well as 10 days after treatment. The biopsies were immediately fixed in 10% neutral buffered formalin for 24 hours, stained with H&E covered, dried and examined microscopically (Carleton et al., 1967).

4) Biochemical analysis:

Blood samples were collected from jugular vein of the examined animals and left for serum separation. Total serum protein were

abscess (Fig. 4). The hair structure never seen above the surface of epidermis. The remaining intra-epidermal part showing invasion with round refractile eosinophylic arthrospores of the causative fungal agent. The underlying dermis was more or less mildly affected with mild focal leukocytic infiltration (Fig. 5).

Concerning the distribution of infection among animals, it was observed that animals of age group from 3 months to 1 year appeared to be more susceptible, followed by animals of age group from 1 year to 2 years and lastly animals of age group from 1 day to 3 months old (Table 1).

Table 1: Incidence of ringworm in relation to age

Age	No. of animals examined	No. of animals infected	Incidence
3 days - 3 M	52	10	19.23
3 M − I year	110	46	41.82
1 year - 2 year	124	36	29.3
Animals>2year	200	15	7.5
Total	486	107	22.01

Table 2: Incidence of ringworm in relation to season

Age | No. of animals | No. of infacted | Levi

Age	No. of animals examined	No. of infected animals	Incidence
Winter	486	53	10.9
Spring	486	36	7.4
Summer	486	12	2,46
Autumn	486	6	1.22

Regarding the seasonality, ringworm was most common in winter (10.9%), followed by spring (7.4%) then summer (2.46%) and lastly Autumn (1.23%).

Electrophoretic pattern of the obtained sera showed variation in the levels of total protein, globulin, Albumin: globulin ratio and between different fraction of the globulins as shown in Table (3).

Table 3: Mean ± SE of total protein and electrophoretic pattern in

Animal Status	Total Protein G/dI	Albumin G/dl	Globulin G/dl	Albumin/ Globulin Ratio	a Globulin G/dl	β Globulin G/dl	y Globulin G/dl
Healthy	7.51±	2.61±	4.9±	0.53±	0.81±	0.65±	3.44±
N= 10	0.64	0.27	0.51	0.022	0.40	0.08	0.236
Infected	6.57 <u>±</u>	2.60 <u>±</u>	3.97 <u>±</u>	0.65±	0.81±	0.63 <u>±</u>	2.53*±
N≈10	0.64	0.24	0.50	0.027	0.13	0.16	0.35

*: Significant at P<0.05

To results of ELISA from measuring serum total immunoglobulins IgG and IgM as shown in Table (4) showed high levels of the total IgG in healthy calves than infected ones.

Table 4: Results of ELISA (mean ± SE of O.D.) for measuring immunoglobulin titers in examined animals.

Animals status	IgG	IgM
Healthy	0.92 ± 0.04	1.6 ± 0.24
Infected	$0.7 \pm 0.03**$	0.91 ± 0.13

^{**:} Sugbufucabt at P<0.001

Treatment of infected animals using tincture iodine 5% needs more time and more application whereas treatment using preparation containing benzoic acid, salicylic acid, and lactic acid in absolute ethanol gave rapid and good clinical and mycological response after only 4 applications one day interval. Moreover, histopathological examination of skin biopsies collected 10 days after application of medicament revealed complete resolution of hyperkeratosis and crusts, the acanthosis previously described have disappeared. The hair started to grow normally. The skin completely returned to normal histological structure (Fig. 6).

DISCUSSION

The hazard of zoonotic infection and reduction of animal price gave ringworm in cattle a major attention.

Incidence of ringworm was recorded in 22.01% of the examined animals. Nearly similar results were recorded by Arslan et al. (1993). Higher incidence was recorded by (Perez-Ruano et al., 1997 and Gyllensvaan, 1999). On the other hand lower incidence was recorded by (Dai et al., 1990; Rashid et al., 1996 and Ammar et al., 1998). The differences in the incidence among studies may be attributed to the degree of environmental contamination in each locality, housing system, hygiene, intensity of animal population and animal susceptibility.

Trichophyton verrucosum was the only dermatophyte detected in this study, in accordance to the results recorded previously by Ammar et al. (1998) and Rhaymah (1999). Records from the farm under study revealed endemic nature and annual occurrence of dermatophytosis especially Trichophyton verrucosum. This observation may be attributed to the survival of dermatophytes spores viable in shed epithelium for many months and even years (Acha and Szyfres, 1989).

- Arslan, S.H.; Al-Kushali, M.N. and Hasso, S.A. (1993): Prevalence of dermatomycosis infection in calves in Mosul. Iraq J. Vet. Sci. 12:1.
- Awad, W.S.A. (1995): Studies on some infectious skin diseases in animals. M.V.Sc. Thesis Fac. Vet. Med., Cairo Univ.
- Carlotti, D.N. and Bensignor, E. (1999): Dermatophytosis due to Microsporum persicolor (13 cases) or Microsporum gypseum (20 cases) in dogs. Vet. Dermatology 10: 17-27.
- Dai, R.L.; Zhao, C.Z.; Lu, Z.X.; Huang, Y.J.; Zhang, Y.D. and Wang, X.L. (1990): Studies on diagnosis and prevention of dermatomycosis in domestic animal: Isolation and identification of Trichophyton verrucosum. Chineese J. Vet. Sci. and Technology, 8: 7-9.
- Edwardson, J. and Andrews, A.H. (1979): An outbreak of ringworm in a group of young cattle. Vet. Rec. 26: 104(21): 474-477.
- Fadlelmula, A.; Agab, H.; Le Horgne, J.M.; Abbas, B. and Abdalla, A.E. (1994): First isolation of Trichophyton verrucosum as the etiology of ringworm in the Sudanese camels (Camelus dromedaries). Rev. Elev. Med. Vet. Pays Trop. 47(2): 184-187.
- Gitao, C.G. (1998): The prevalence of dermatophilus congolensis infection of camels in four rearing areas in Kenya and the presence of a mixed infection with Trichophyton verrucosum. Israel J. Vet. Mcd. 53(3): 89-93.
- Goslen, J.B. and Kobayashi, G.S. (1987): Mycological infection. In: Fitzpatrick, T.B.; Eisen, A.Z., Wolf, K. Ed. Dermatology in general medicine. 3rd ed. New York Macgraw. Hill pp. 2201-2229.
- Gyllensvaan, C. (1999): A survey of ringworm in the winter of 1998. Svensk Veterivartidning 51(4): 189-192.
- Haab, C.; Bertschinger, H.U. and Von Rotz, A. (1994): Epidemiology of trichophytosis in fattening calves in regard to the prevention of leather defects. Schweiz Arch Tierheilk 136(6-7): 217-226.
- Haggag, Y.: Draz, A. and Samaha, H. (1999): Soil as a reservoir of certain dermatophytes and other fungi to man and animals. Alex. J. Vet. Sci. 15(1): 1-9.
- Hungerfords, T.C. (1990): Diseases of livestock, 9th edition Bibliography, Pp. 542-543.
- Job, K.; Rennedy, P.C. and Palmer, N. (1993): Dermatophytosis in pathology of domestic animals 4th Ed. Pp 661.

recorded in winter and spring. This finding agrees to that reported previously by (Fadlelmula et al., 1994 and Ammar et al., 1998) who recorded a high prevalence in July and a minumum prevalence in March.

Serum biochemical analysis revealed non significant alteration in total serum protein, albumin, alpha and beta fractions of globulin between the infected and clinically healthy animals. In contrast gamma globulin fraction showed a significant increase in clinically healthy ones. In addition, the total serum immunglobulins (Ig) G were significantly higher in healthy animals than in infected ones whereas the total immunoglobulins (Ig) M showed non significant differences. The significant decrease in gammaglobulin in infected animals than normal one indicated that the immue reponse against ringworm are mainly occurred via cell mediated immune response (Koed, 1997 and Lund et al., 2001).

The preparation containing 2.5%, Benzoic 2.5 % Salicylic acid and 2% lactic acid in absolute ethanol offer efficient result in eliminating the infection with 100 recovery rate after only 4 application one day interval. The efficacy of the preparation is due to the acidity as well as the antifungal activity of its components (Perscott et al., 2000 and Maddison et al., 2002). In addition examination of skin biopsies taken 10 days after treatment confirm the efficacy of the medicament in returning the skin to its normal histological structure.

REFERENCES

Abdel-Halim, M; Youssef, H. Ramadan, S.A. and Refai, M. (1988):
Dermatophytosis in Egyptian sheep and goats. Vet. Med. J. 36:
1010-1016.

Abou Zaid, A.A. (1996): Studies on some skin affection in cattle. 7th Sci. Cong. 17-19 Nov., Fac. Vet. Med., Assiut Egypt.

Abu-Gabal, M.; El-Gelil, G.A.; El-Nor, E.A.; El-Rehim, D.A. (1976):
Animal ringworm in Upper Egypt. Sabouraudia 14(1): 33-36.

Acha, P.N. and Szyfres, B. (1989): Zoonoses and communicable diseases common to man and animals. 2nd ed. Pan American Health Organization, Washington, USA.

Ammar, K.M.; Al-Gaabry, M.H.: Nassif, M.M. and Foad, F.M. 91998): Epidemiological studies and therapeutic trials of dermatophytosis in cattle and sheep. Eight Scientific Congress Fac. Vet. Med. Assiut Univ., 15-17 November 1998. Page 708-717.

- Arslan, S.H.; Al-Kushali, M.N. and Hasso, S.A. (1993): Prevalence of dermatomycosis infection in calves in Mosul. Iraq J. Vet. Sci. 12:1.
- Awad, W.S.A. (1995): Studies on some infectious skin diseases in animals. M.V.Sc. Thesis Fac. Vet. Med., Cairo Univ.
- Carlotti, D.N. and Bensignor, E. (1999): Dermatophytosis due to Microsporum persicolor (13 cases) or Microsporum gypseum (20 cases) in dogs. Vet. Dermatology 10: 17-27.
- Dai, R.L.; Zhao, C.Z.; Lu, Z.X.; Huang, Y.J.; Zhang, Y.D. and Wang, X.L. (1990): Studies on diagnosis and prevention of dermatomycosis in domestic animal: Isolation and identification of Trichophyton verrucosum. Chineese J. Vet. Sci. and Technology, 8: 7-9.
- Edwardson, J. and Andrews, A.H. (1979): An outbreak of ringworm in a group of young cattle. Vet. Rec. 26: 104(21): 474-477.
- Fadlelmula, A.; Agab, H.; Le Horgne, J.M.; Abbas, B. and Abdalla, A.E. (1994): First isolation of Trichophyton verrucosum as the etiology of ringworm in the Sudanese camels (Camelus dromedaries). Rev. Elev. Med. Vet. Pays Trop. 47(2): 184-187.
- Gitao, C.G. (1998): The prevalence of dermatophilus congolensis infection of camels in four rearing areas in Kenya and the presence of a mixed infection with Trichophyton verrucosum. Israel J. Vet. Mcd. 53(3): 89-93.
- Goslen, J.B. and Kobayashi, G.S. (1987): Mycological infection. In: Fitzpatrick, T.B.; Eisen, A.Z., Wolf, K. Ed. Dermatology in general medicine. 3rd ed. New York Macgraw. Hill pp. 2201-2229.
- Gyllensvaan, C. (1999): A survey of ringworm in the winter of 1998. Svensk Veterivartidning 51(4): 189-192.
- Haab, C.; Bertschinger, H.U. and Von Rotz, A. (1994): Epidemiology of trichophytosis in fattening calves in regard to the prevention of leather defects. Schweiz Arch Tierheilk 136(6-7): 217-226.
- Haggag, Y.: Draz, A. and Samaha, H. (1999): Soil as a reservoir of certain dermatophytes and other fungi to man and animals. Alex. J. Vet. Sci. 15(1): 1-9.
- Hungerfords, T.C. (1990): Diseases of livestock, 9th edition Bibliography, Pp. 542-543.
- Job, K.; Rennedy, P.C. and Palmer, N. (1993): Dermatophytosis in pathology of domestic animals 4th Ed. Pp 661.

- Kelly, W.R. (1984): Veterinary Clinical Diagnosis. 3rd ed. Bailliera Tindal London.
- Koed, N.P. (1997): Vaccination of unweaned calves against ringworm. Dansk Veterinaertidsskrift. 80(16): 695-699.
- Koneman, E.W. and Roberts, G.D. (1985): Practical laboratory mycology. 3rd edition. Williams and wilkins. Baltimore, M.D., 1985.
- Laemmii, A.R. (1970): Cleavage of structural proteins during assembly of the head of bacteriophage. T4. Nature, 227: 600-685.
- Lund, A.; Bratberg, A.M.; Solbak, I.T. (2001): In vitro release of interferone gamma by trichophyton stimulated whole blood cell cultures from ringworm vaccinated and conrol calves experimentally inoculated with Trichophyton verrucosum. Vet. Dermatology. 12(2): 75-80.
- Maddison, J.; Page, S. and Church, D. (2002): Small Animal Pharmacology. 1st ed. Harcourt pupilisher limited. Pp. 539.
- Markus, R.; Huzaira, M.; Anderson, R.R. and Gonzalez, S. (2001): A better potassium hydroxide preparation. In vivo diagnosis of tinea with Cofocal microscopy. Arch Dermatology 137(8): 1076-1078.
- Midgley, G.; Clayton, Y. and Hay, R.J. (1997): Medical Mycology. Mosby-Wolf Chicago.
- Nashwa, Khalifa, O. (2001): Zoonotic aspect of Trichophyton mentagrophytes in rabbit farms. Beni Suef Vet. Med. J. 11(2-A): 49-56.
- Pal, M.; Sejra, A.P. and Rao, N.M. (1995); Prevalence of dermatophytosis in water buffaloes (Bubalus bubalis). Buffalo Bulletin. 14(3): 51-54.
- Pandey, V.S. and Cabaret, J. (1980): The distribution of ringworm lesions in cattle naturally infected by Trichophyton verrucosum Ann. Rec. Vet. 11(2): 179-184.
- Pepin, G.A. and Oxenham, M. (1986): Zoonotic dermatophytosis (ringworm). Vet. Rec. 118: 110-111.
- Perez-Ruano, M.; Abeledo, M.A.; Gonzalez, M. and Medoza, A. (1997): Vaccine against bovine trichophytosis. III. Evaluation. Revista de Salud Animal. 19(2): 85-88.
- Prescott, J.F.; Baggot, J.D. and Walker, R.D. (2000): Antimicrobial therapy in Veterinary Medicine. 3rd ed. Iowa state University Press. Blackwell scientific puplication.

Radostits, O.M.; Blood, D.C. and Gay, C.C. (1994): Veterinary Medicine. 8th edition. Bailliere, London, U.K.

Rafaa, A.H.A.M. (1992): Studies on some mycotic affections in animals.

M.V.Sc. Fac. Vet. Med. Cairo Univ.

Rashid, M.A.; Noorddin, M. and Dey, A.S. (1996): Distribution of dermatomycosis in cattle, goats and in contact human. Bangladesh Veterinarian. 13(1-2): 9-12.

Rhaymah, M.S. (1999): Clinical and therapcutic study of ringworm in bull calves. Iraq J. Vet. Sci. 12:1.

SAS (1987): Statistical Analysis. User's Guide: Statistics, SAS Institute Cary, North Carolina.

Scott, D.W.; Miller, W.H. and Griffin, O.E. (1995); Muller and Kirks small animal dermatology 5th ed. Philadelphia: W.B. Saunders 1995.

Scott, D.W. (1988): Large animal dermatology. Saunders, London.

Seddik, A.; Abdel Kader, H.A. and El-Shanawany, A.A. (1994):
Dermatophytes and other fungi isolated from sheep in Assiut
Province, Assiut Vet. Med. J. 30: 6.

Sharma, S.R.; Dakshinkar, N.P.; Shoot, V.M. and Sapre, V.A. (1994): evaluation of crude extract of garlic in dermatophytosis of calves. Inter. J. Anim. Sci. 9(2): 239-240.

Singh, N.; Yadav, J.S.; Singh, A.P. and Sharma, S.N. (1997):
Clinicoepidemiological studies on bovine dermatiophytosis in and around Bikaner. Ind. J. Anim. Sci. 67(10): 845-848.

Stevenson, W.J. and Hughes, K.L. (1988): Synopsis of zoonosis (2nd edition) Ringworm. Australian Government Publishing Services. Canberra, PP. 99-106.

St-Germain, G. and Summerbell, R. (1996): Identifying filamentous fungi. Star Publishinh. Belmont, C.A., 1996.

Tegami, H. et al. (1982): Analysis of trans epidermal leucocytic chemotaxis in experimental dermatophytosis in guinea pigs. Ach. Dermatol. Res. 273: 205-217) cited after Jobe et al., 1993).

Voller, A.; Billwell, D. and Bartlett, A. (1976): Microplte enzyme immunoassays for the immunodiagnosis of virus infections. In: Manual of clinical immunology. PP. 506 Washington: Ajmerican Society of Microbiology.

Wabacha, J.K.; Gitau, G.K.; Bebora, L.C.; Bwanga, C.O.; Wamuri, Z.M. and Mbithi, P.M.E. (1998): Occurrence of dermatomycosis (ringworm) due to Trichophyton verrucosum in dairy calves and its spread to animal attendants. J.S. Afr. Vet. Assoc. 69(4): 172-173.

Vet. Assoc. 69(4): 172-173.

Weichselbum, T.E. (1946): An accurate and rapid method for determination of proteins in small amounts of blood scrum and plasma. Am. J. of Chem. Pathol. 10: 40-45.

Wright, A.J. (1989): Ringworm in dogs and cats. J. Smal Animal Practice. 30: 242-249.





Fig.1. Lesions of ringworm in Friesian calves.

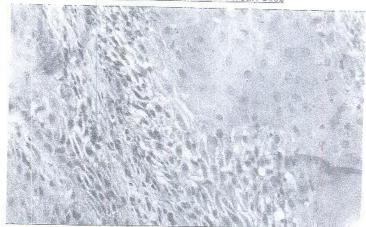


Fig.2.Epidermis showing swelling of spines of brickle cell layers with marked acanthosis. H&E (X400).

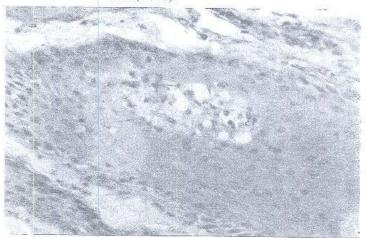


Fig. Epidermis of calf infected with T. verrucosum showing intra epidermal vesicles formations. H&E (X400).

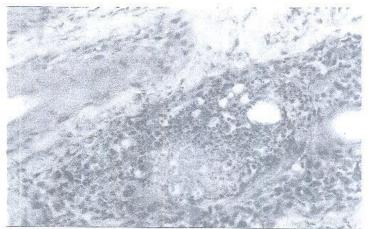
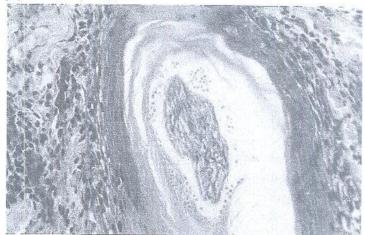


Fig.4. Epidermis of calf infected with T, verrucosum showing intra epidermal abscess formation and focal infiltration of inflammatory cells in dermal papillae. H&E (X400).



Pig.5. Epidermis of calf infected with T, verrucosum showing presence of arthrospores within destroyed hair with focal leukocytic infiltration in dermis. H&E (X400).

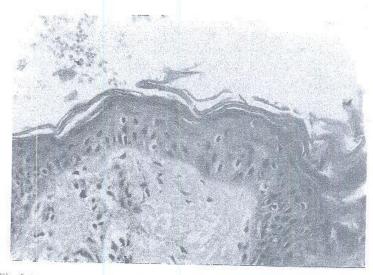


Fig.6. Normal histological structure of skin biopsies after treatment. H & E (X400).