

Amyloidosis in lymphoid organs of buffaloes

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ABSTRACT

A total of 50 spleens and 250 lymph nodes collected from buffaloes were examined, in which amyloid deposition was identified in 3 spleens and 144 lymph nodes, included 30 prescapular, 38 prefemoral, 29 hepatic, 23 mesenteric and 24 bronchial lymph nodes. Cytological examination revealed the presence of amorphous, homogenous, extracellular material suggestive of amyloid along with associated cellular changes. Histopathological examination demonstrated eosinophilic, amorphous extracellular deposits within the lymphoid tissues, lead to partial distortion of the normal architecture of the affected organs. The findings of the present study indicated that increased occurrence of amyloidosis in the lymphoid organs of buffaloes and highlighting the significance of cytological and histopathological techniques in the detection of this condition.

Keywords: Amyloidosis, Buffaloes, Cytology, Histopathology, Lymphoid organs

INTRODUCTION

Amyloidosis is a pathological condition characterized by extracellular deposition of insoluble fibrillar proteins known as amyloid in various tissues and organs. These amyloid fibrils are composed of misfolded protein subunits arranged in a Beta-pleated sheet configuration, which accumulate in the extracellular spaces and disrupt normal tissue architecture and function^{1,2}. Depending upon the distribution of amyloid deposits, the condition may occur as localized or systemic amyloidosis.

Amyloidosis was reported in a wide range of animal species viz., cattle, buffaloes, dogs, cats and birds, affecting organs such as the liver, spleen, kidneys and lymph nodes^{2,3}. Deposition of amyloid within these organs may leading to compression and atrophy of adjacent cells and ultimately impair organ function. Lymphoid organs such as the spleen and lymph nodes play a critical role in immune responses and are frequently examined during pathological investigations. Amyloid deposition in these tissues may alter the normal lymphoid architecture and interfere with immunological functions². Although amyloidosis was documented in several organs of animals, reports describing its occurrence in lymphoid tissues of buffaloes were limited.

Therefore, the present study was undertaken to investigate the occurrence of amyloidosis in the lymphoid organs of buffaloes through cytological and histopathological examination.

MATERIALS AND METHODS

A total of 50 spleens and 250 lymph nodes including prescapular, prefemoral, hepatic, mesenteric, bronchial lymph nodes were collected from buffaloes slaughtered in slaughterhouses in and around Tirupati and examined for the present study with approval obtained from the Institutional Animal Ethics Committee (IAEC) wide no: - 281/ go/ ReBi/ S/ 2000/ CPCSEA/ CVSc/ TPTY/ 047/ Veterinary Pathology/ 2025 dated: 02-08-2025.

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The impression smears were prepared from the collected spleens and lymph node samples using standard preparation techniques. The air-dried smears were stained with Wright's stain and Leishman's stain and examined under a light microscope⁴.

Representative tissue samples of spleen and lymph nodes were fixed in 10% neutral buffered formalin, processed by routine paraffin embedding technique, and sections of approximately 4-5 microns thickness were prepared using a rotary microtome. The sections were stained with hematoxylin and eosin (H&E) for routine histopathological examination^{4,5}.

To confirm the presence of amyloid, selected tissue sections were subjected to Congo red and

crystal violet staining techniques. The stained sections were examined microscopically for characteristic staining reactions indicative of amyloid deposition^{4,6}.

RESULTS

Histopathological sections revealed amyloid deposition in 3 (6%) out of 50 spleens and 144 (57.6%) out of 250 lymph nodes examined. The overall occurrence of amyloidosis in the present study was 49% of lymphoid organs of buffaloes (Table.1).

Cytological smears of spleen and lymph nodes revealed the presence of amorphous, homogenous, extracellular material suggestive of amyloid. The material appeared as pale to eosinophilic, structureless deposits distributed

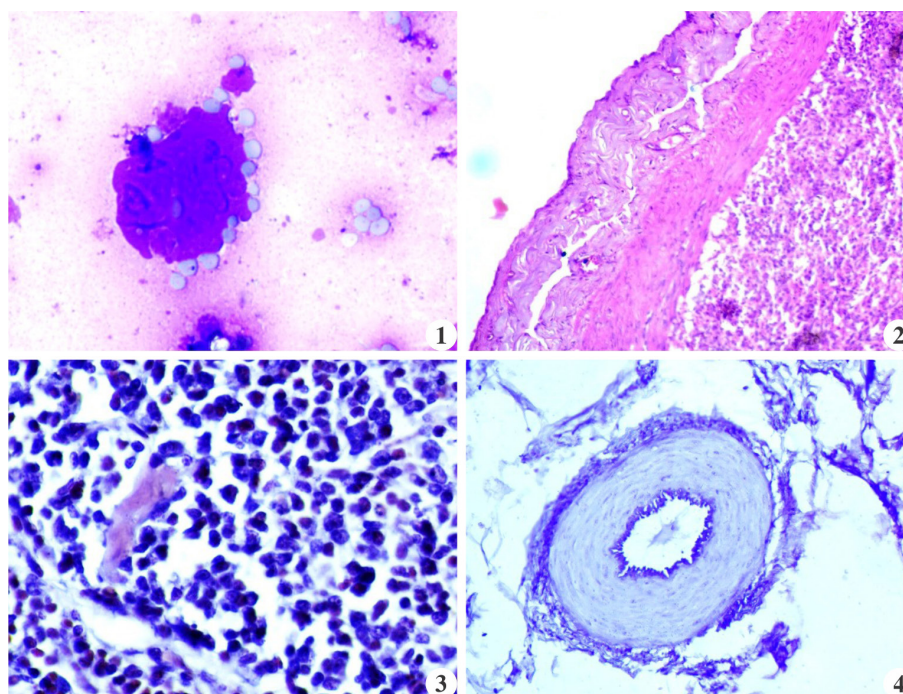
among the cellular elements of the lymphoid tissue (Fig.1).

In the spleen, amyloid deposits were observed as extracellular, eosinophilic, amorphous and hyalinized deposits within the capsule (Fig.2), red pulp and were distributed around the sheathed arteries. In addition deposition of amyloid was also noted in the capsule and trabeculae, resulting in partial alteration of the normal splenic architecture.

Among the lymph nodes examined, amyloid deposition was recorded in 30 (60%) prescapular, 38 (76%) prefemoral, 29 (58%) hepatic, 23 (46%) mesenteric and 24 (48%) bronchial lymph nodes. Histologically, the deposits were observed in the hilus, around blood vessels (Fig.3), lymphatic vessels and, lymphoid follicles (Fig.4).

Table.1. Distribution of amyloid deposition in spleen and lymph nodes of buffaloes

Lymphoid tissues	Number of organs examined	Number of positive cases	Percentage (%)
1. Spleen	50	3	6.0
2. Prescapular lymph node	50	30	60.0
3. Prefemoral lymph node	50	38	76.0
4. Hepatic lymph node	50	29	58.0
5. Mesenteric lymph node	50	23	46.0
6. Bronchial lymph node	50	24	48.0
Total	300	147	49.0



Amyloidosis in lymphoid organs in buffaloes. Fig.1. Amorphous, homogenous, extracellular material - Leishman's stain - spleen x400; Fig.2. Extracellular, eosinophilic, amorphous and hyalinized deposits within the capsule - H&E - spleen x100; Fig.3. Red coloured deposits of amyloid in the lymphoid follicles - Congo red stain - prescapular lymph node x400; Fig.4. Dark purple coloured deposits of amyloid in and around blood vessels - Crystal violet stain - prefemoral lymph node x100.

Follicular amyloidosis was predominantly noticed in the hepatic and mesenteric lymph nodes, where amyloid deposits were mainly localized within the lymphoid follicles.

The presence of amyloid was further confirmed by Congo red stain in which amyloid appeared as red colored deposits and in Crystal violet stain as dark purple colored deposits in the spleen and lymph nodes.

DISCUSSION

The prevalence was lower compared to the findings of nodular ('sago' spleen) type of splenic amyloidosis (5.3%) in cattle. A higher prevalence was documented in sheltered cats, where systemic amyloidosis with splenic involvement was recorded in 52-73% of animals, and in captive cheetahs where splenic amyloid was frequently encountered as a part of systemic disease⁸. Histopathologically, amyloid deposits appeared as homogenous eosinophilic extracellular material, which is considered a characteristic microscopic feature of amyloidosis^{2,3}. In domestic ruminants such as cattle and sheep, splenic amyloidosis was described qualitatively as an occasional or nodular ('sago' spleen) lesion rather than a common finding³. It might be attributed to fewer chronic inflammatory conditions, reduced antigenic stimulation, or species variation in susceptibility to systemic amyloidosis². Similar patterns of amyloid distribution in the spleen was reported in domestic animals, where deposits commonly occur in association with vascular and connective structures of the splenic parenchyma^{2,3}. Amyloidosis in animals were reported in several organs including the liver, spleen, kidneys and lymph nodes, indicating that the disease frequently occurs as a systemic condition affecting multiple tissues⁹. In the liver, amyloid deposition was described in the space of Disse and along hepatic sinusoids, lead to compression and atrophy of hepatocytes². Similarly, in the kidneys, amyloid deposits were commonly found in the glomeruli and interstitial tissues, which might result in renal dysfunction³.¹⁰ described renal amyloidosis in cattle confirmed by Congo red staining, while¹¹ reported amyloid deposition associated with chronic inflammatory diseases in cattle. Continuous immune stimulation within lymph nodes during chronic infections, inflammatory diseases or immune-mediated conditions could therefore result in amyloid accumulation within lymphoid follicles and vascular walls^{2,12}. Comparable localization of amyloid in lymphoid follicles and vascular structures of lymph nodes was described in animals affected with systemic amyloidosis^{2,9}. Similar cytomorphological appearance of amyloid was described in cytological preparations of affected tissues, where it appeared as homogenous extracellular deposits¹³. Confirmation of amyloid deposition in the present study was achieved with special

staining techniques. Congo red stain demonstrated red deposits, while crystal violet stain revealed dark purple deposits, which were considered characteristic reactions for amyloid material^{4,6}. The development of amyloidosis in animals is commonly associated with chronic inflammatory or infectious conditions that stimulate prolonged hepatic production of serum amyloid A protein, an acute-phase reactant. Persistent elevation of this protein leads to its misfolding and conversion into insoluble amyloid fibrils that accumulate in tissues such as the spleen, liver, kidney and lymph nodes^{2,3,9}.

CONCLUSION

The present study demonstrated the increased occurrence of amyloidosis in the lymphoid organs of buffaloes. Amyloid deposition was successfully detected through cytological and histopathological examination and confirmed by special staining techniques, highlighting the importance of these methods in the identification of amyloidosis in buffalo lymphoid tissues.

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Conflicts of Interest: None

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