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SECONDARY AMYLOIDOSIS IN DALL'S SHEEP

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Spontaneous amyloidosis is a naturally occurring disease complex that has been described in more than 30 mammalian species (Jakob, 1971, Vet. Pathol. 8: 292-306).

Although amyloidosis may develop in the absence of any other recognizable disease process, its appearance often is preceded by long-standing inflammatory disorders. These observations gave rise to the concept of primary and secondary amyloidosis, the former tending to localize in the cardiovascular system and the latter tending to focus on the kidneys, liver, spleen, and adrenals (Glenner, 1980, N. Engl. J. Med. 302: 1283-1292).

Amyloid, a pathologic proteinaceous substance deposited extracellularly in tissues, has typical staining properties with commonly used dyes and shows green birefringence with Congo-red staining and polarized light examination (Glenner, 1976, Int. Rev. Exp. Pathol. 15: 1-92). Recent advances in biochemical and immunologic analyses of human amyloid have disclosed the presence of at least five different types of amyloid proteins. Amyloid of secondary amyloidosis is designated protein AA (Fujihara, 1981, Lab. Invest. 44: 55-60).

A relatively simple histochemical method, based on the affinity of amyloid for Congo red dye after exposure to potassium permanganate, is available to distinguish different chemical types of amyloid. With this method secondary amyloid is sensitive to permanganate treatment in that it loses its Congo red affinity and polarization characteristics while other classes of amyloid are resistant to permanganate treatment (Wright, 1977, Lab. Invest. 36: 274-281).

This report describes naturally-occurring secondary amyloidosis in seven captive Dall's sheep *(Ovis dalli dalli)* that were maintained in a zoological park.

Seven of 25 adult Dall's sheep ranging from 5 to 10 yr old died over a period of 7 mo. Necropsy showed that five of the seven sheep had severe chronic pneumonia. Microscopic diagnoses varied but included chronic pneumonia of various types in six sheep, chronic interstitial nephritis in two, and amyloidosis in all. Inflammatory conditions, generally chronic, accompanied the amyloidosis in all sheep.

Tissues from these seven sheep were fixed in 10% formalin, routinely processed, and stained with hematoxylin and eosin (H & E). In addition, the following special stains were used to identify and characterize the amyloid deposits: Congo red with and without polarized light examination, crystal violet, thioflavin T, and pretreatment of histological sections with potassium permanganate before Congo red staining to distinguish secondary amyloid. Positive controls were used with the special stains.

Table 1 lists the results of the staining procedures used. In all seven sheep the various amyloid-containing tissues reacted similarly. A typical hyaline appearance was evident in H&E sections while Congo red-stained amyloid showed typical orange-salmon color with bright-field microscopy and green birefringence with polarized light. Amyloid appeared purplish violet with crystal violet and with thioflavin T showed white fluorescence in ultraviolet light. In contrast, tissue sections pretreated with potassium permanganate and then

TABLE 1. Histochemical characteristics of amyloid in Dall's sheep affected with secondary amyloidosis.

Sheep No.	Tissue	H&E	Congo Red	Congo Red Polarized	pp[a] and Congo Red	pp and Congo red Polarized	Crystal Violet	Thioflavin T
1	Kidney	+	+	++	-	-	+	+
	Liver	±	+	+	-	-	+	++
2	Kidney	+	+	+	-	-	+	+
	Liver	++	+	++	-	-	+	+
	Adrenal	+	+	+	-	-	±	+
3	Kidney	+	NA	NA	NA	NA	NA	NA
	Liver	+	NA	NA	NA	NA	NA	NA
4	Liver	+	±	+	-	-	+	+
	Kidney	+	NA	NA	NA	NA	NA	NA
5	Kidney	+	+	+	-	-	+	++
	Liver	+	+	+	±	-	+	+
	Adrenal	±	+	+	±	-	+	+
	Lymph node	+	+	+	-	-	+	+
	Spleen	++	+	+	-	-	+	+
6	Kidney	+	+	++	-	-	+	+
	Liver	+	+	++	-	-	+	+
	Heart	+	±	+	-	-	+	+
	Adrenal	+	+	+	-	-	+	+
	Spleen	+	+	+	-	-	+	+
7	Kidney	+	+	+	-	-	+	++
	Adrenal	+	+	±	-	-	+	++

++ = positive, strong staining, birefringence or fluorescence
\+ = positive staining, birefringence or fluorescence
± = equivocal staining, birefringence or fluorescence
\- = negative staining, birefringence or fluorescence
a = pretreatment with potassium permanganate
NA = tissue not available

stained with Congo red generally did not show the typical findings when examined with bright field and polarized light microscopy. The amyloid lost affinity for Congo red dye and its birefringence was totally obliterated. These results indicated that in all seven sheep the amyloid deposits consisted of protein AA or secondary amyloid.

Information on amyloidosis in wild mammals is minimal and is restricted to a few case reports. A review of spontaneous amyloidosis in mammals cites reports of amyloidosis in various species of wild mammals but generally the number of cases reported is small (Jakob, op. cit.).

There are two reports of amyloidosis in Rocky Mountain bighorn sheep *(Ovis canadensis canadensis).* The first described amyloidosis in three of seven wild adults obtained over a 9-yr period. In each, amyloid was deposited in various parenchymatous organs and there was an associated chronic inflammatory condition (Hadlow, 1962, J. Am. Vet. Med. Assoc. 141: 243-247). The second report concerned a captive herd in which 17 sheep died after varying periods of captivity (11-865 days). The primary pathologic finding in 16 sheep was bronchopneumonia. Amyloidosis was observed in seven of the 17 (41%) sheep (Wolfe, 1973, J. Wildl. Dis. 9: 12-17). In neither of these reports was the specific chemical type of amyloid determined.

The prevalence of secondary amyloidosis in wild mammals is unknown and its importance as a disease is yet to be determined. Available evidence indicates, however, that secondary amyloidosis may be a specific and important complication in wild sheep.

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