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'Shrivelled': a hereditary degeneration of the lens in the house mouse

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This paper reports preliminary studies on a gene-determined degeneration of the ocular lens in the house mouse. The term 'Shrivelled' (gene symbol *Svl*) is proposed for the mutant, since this describes a characteristic appearance of the lens during the degenerative process.

MATERIALS AND METHODS

The progress of the condition was studied by observations (1) of unanaesthetized animals through a dissecting microscope; if the eyes were pigmented the iris was dilated with homatropine; (2) of gross dissection of eyes fixed in Bouin's solution; (3) of histological sections of eyes fixed in Bouin's solution, embedded and cut in celloidin and stained with Harris' ammonium alum hematoxylin and eosin.

INHERITANCE

The mutant condition was first noted by Dr L. Pinsky in a shipment of mice of the inbred A/Jax strain received from the Jackson Laboratory, Bar Harbor, Maine. One animal had a dense white opacity of the lens in both eyes, and other lenticular abnormalities to be described below. No other mouse with this condition has been observed in the Jackson Laboratory A/Jax colony, according to Dr Margaret Green. The original mutant female was mated to unaffected males and produced three litters, with a total of 16 offspring (8♂, 8♀) all of which showed the peculiar degenerative process of the lens which characterizes the Shrivelled phenotype. None of these animals developed the marked opacities seen in their mother. If (as will be demonstrated below) the condition is caused by a single gene, with intermediate dominance, the original female was, therefore, almost certainly homozygous for it. For the following genetic analysis, animals were classified as homozygotes if they developed cataract (i.e. a more or less dense, white opacity of the lens) within the first six weeks of life, and showed a rapid progression of the lens degeneration. Six mice so classified, mated to unaffected animals, have had a total of 81 offspring all showing the Shrivelled phenotype without early cataract formation, which suggests that the classification is valid.

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Table 1 presents the results of crossing heterozygous Shrivelled mice *inter se*, or to unaffected animals. Since no differences in segregation ratios were noted between crosses within the A/Jax strain and outcrosses the results have been combined. The occurrence of Shrivelled in both male and female offspring of heterozygous Shrivelled females outcrossed to unrelated males ruled out sex-linked and autosomal recessive inheritance, and supported the hypothesis of autosomal dominant inheritance. Table 1, line 3, shows that matings of heterozygous 'Shrivelled' with unaffected animals resulted in a ratio of 132*Svl*: 143 + offspring. This does not differ significantly from the 1:1 ratio expected on the basis of a single gene segregation ($\chi^2 = 0{\cdot}441$, $p > 0{\cdot}5$). Table 1, line 2, rules out the possibility of a completely sex-linked (either X or Y) dominant gene since affected males and females occur in the offspring of affected males crossed to unaffected females.

Table 1. *Segregation of Shrivelled* (Svl) *in various crosses*

		Svl/*Svl*		*Svl*/+		+/+		T
♀	♂	♂	♀	♂	♀	♂	♀	
1. *Svl*/+	+/+	0	0	26	42	34	37	139
2. +/+	*Svl*/+	0	0	29	35	41	31	136
3. Sum of crosses 1 and 2		0	0	55	77	75	68	275
4. *Svl*/+	*Svl*/+	15	18	40	33	16	15	137
*5. +/+	*Svl*/+ with *Svl* mother	0	0	9	12	14	12	47
*6. +/+	*Svl*/+ with *Svl* father	0	0	12	16	17	15	60

* Extracted from line 2.

Matings between heterozygous animals resulted in a ratio of 33 homozygous Shrivelled to 73 heterozygous Shrivelled to 31 non-Shrivelled offspring. This does not differ from the expected 1:2:1 ratio for a single autosomal gene ($\chi^2 = 0{\cdot}650$, $p > 0{\cdot}7$). There is no suggestion of a difference in sex ratio of affected offspring from *Svl*/+ males outcrossed to normal females, when classified according to whether the male received his *Svl* gene from his mother (Table 1, line 5) or his father (line 6). Thus there is no evidence of partial sex-linkage.

To test whether *Svl* was allelic to the recessive gene 'lens rupture' (Fraser & Herer, 1950), which also causes cataracts, though of a different type, heterozygous Shrivelled animals were crossed to homozygous lens rupture animals. Female offspring with the Shrivelled phenotype were backcrossed to lens rupture males. Of 25 offspring, 10 were neither lens rupture nor Shrivelled, demonstrating that the two mutants are not alleles. There is some indication, however, that the Shrivelled condition may suppress the expression of the lens rupture phenotype, and this will be investigated further.

Preliminary linkage tests (involving about 50 backcross animals in each case) showed no evidence of linkage of *Svl* with *a*, *b*, *c*, or *N*.

It is concluded that the Shrivelled condition is caused by an autosomal gene, of intermediate dominance, with high penetrance.

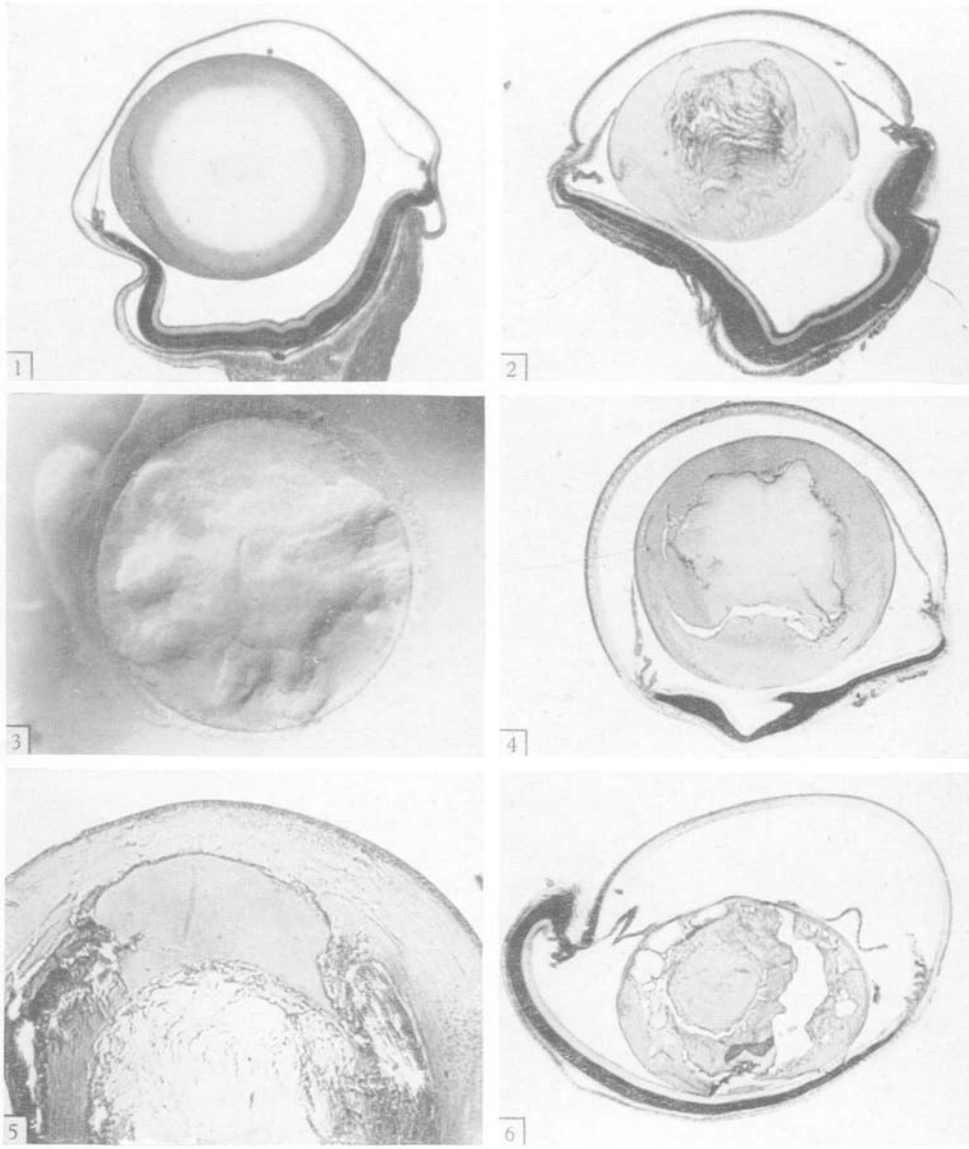

Fig. 1. A section of an eye from a 9-week old albino mouse.

FIG. 2. A section of the eye from a 9-week-old heterozygous *Svl* mouse at the 'pepper' stage. Note the areas of amorphous material among the fibres of the cortex. The irregularities of the retina and choroid are artefacts.

FIG. 3. A fresh dissection of an eye from a heterozygous *Svl* mouse at the 'pseudopodial' stage. The irregular central body can be seen more clearly in the photograph than through the microscope, where it can only be defined by its refractive outline.

FIG. 4. A section of the eye from a 9-week-old heterozygous *Svl* mouse at the 'pseudopodial' stage.

FIG. 5. A high-power view of an area of amorphous degenerated lens fibre material near the anterior pole in a 16-week-old heterozygous *Svl* mouse.

FIG. 6. A section of an eye from a homozygous *Svl* mouse about 60 weeks old. Note the shrunken, vacuolated lens.

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DESCRIPTION

Heterozygote

By *in vivo* observation, the eye of the unaffected albino mouse appears entirely clear. The eyes of 6 Svl mice observed at 12 days of age appeared normal. During the third week of life, a refractile body, about one-half to three-quarters the size of the normal lens, can be seen in the central region of the eye. It may be spherical, or more often irregular in shape, and is usually transparent. Gross dissection of unfixed eyes shows that the lens is normal in size and shape, and that the body observed *in vivo* is not the whole lens, but is centrally located within it. At this age, *in vivo* observations also often reveal fine lines 'etched' on the anterior surface of the lens. On the yellow surface of the fixed lens these appear as narrow brown bands, radiating from the anterior pole.

Histologically, the fibres at the periphery of the lens nucleus are swollen and appear to be degenerating into a homogeneous eosinophilic material. The nuclei of the fibres near the area of degeneration are swollen, and those immediately adjacent to it break up into numerous deeply staining granules. The homogeneous material at some points approaches the lens capsule anteriorly (Fig. 2), and the interface at the area of contact appears to be the basis for the fine 'etched' lines observed *in vivo*.

By the 5th or 6th week the central body as seen *in vivo* usually has a characteristic shape resembling that of a pepper, viewed end on. There are rounded ridges running perpendicular to the equator and taking their origin from the anterior pole. This appearance has been seen as late as the 10th week, but is more common in the 6th or 7th weeks. The central body may be clear or cloudy and, since the periphery of the lens cannot be seen without special optical equipment, the lens appears to be decreased in size—that is, shrivelled. Dissections of unfixed eyes again demonstrate that the lens is normal in size and shape, and clear peripherally, with the Shrivelled central body clearly visible within it. In some cases, at the 6th week the shape of the central body may be very irregular, but occasionally it may still appear round. The surface of the lens may still appear etched.

The 'pepper' stage is often followed by a stage described as 'pseudopodial' in which the ridges have become larger and more irregular (Fig. 3). The central body may be cloudy or opaque posteriorly, or clear peripherally with a cloudy central area, or entirely clear. In a few cases its outline is indistinct. This stage may be seen at the 7th week to the 12th week, but was most often seen from the 7th to 10th week. Around the 10th week the lens occasionally develops small, dense, discrete opactities especially towards the posterior pole.

At the 'pepper' and 'pseudopodial' stages, histologically, the lens nucleus appears irregular and the cortex contains large amounts of amorphous material adjacent to more peripheral normal lens fibres (Figs. 4 and 5). In dissection of fixed lenses the outer fibres can be easily peeled off the central body, revealing it as a homogeneous mass with peripheral projections. On cutting, it has the consistency of hard paraffin wax, with no trace of the fibrous structure still apparent in the peripheral cortex.

After about the 11th week, the central body gradually becomes relatively smooth and spherical, the lens usually becomes progressively more cloudy, and the nucleus tends to become more opaque, although there is a great deal of variability, and lenses with no opacities have been seen as late as 36 weeks of age. Histologically, there is progressive involvement of the peripheral cortex, with loss of fibre structure, and degeneration of fibre nuclei. The amorphous material may appear as numerous small droplets, and the nucleus may be displaced posteriorly. Clear vacuoles may develop in the cortex. In older animals there may be degenerative changes in the capsule, perhaps with adhesions to the iris, but no pathological changes have been observed in the retina, cornea, ciliary body, or sclera.

Homozygotes

In the 3rd or 4th week of life the presumed homozygotes have a cloudy lens, with an irregular central body within it that usually contains small irregular iridescent or opaque specks, and a dense white body of the size and location of the nucleus. As early as 40 days there may be opacities in the cortex as well, and the whole lens becomes progressively smaller and more opaque. Histologically, by about 6 months the lens is small, markedly vacuolated, particularly around the equator, and there are very few recognizable fibres left. Most of the lens substance consists of a mixture of eosinophilic amorphous and granular material (Fig. 6).

Apart from the eye, no abnormalities have been observed in either homozygous or heterozygous Shrivelled animals.

DISCUSSION

The Shrivelled phenotype appears to be caused by a mutant gene which produces an unusual degenerative process in the ocular lens. Since the first mutant animal observed was from the highly inbred A/Jax line and was very probably homozygous for the mutant gene, it is likely that the mutant may, by now, exist in several other A/Jax colonies besides our own.

The genetic analysis shows that the Shrivelled condition is caused by a single autosomal gene, with intermediate dominance, since lens degeneration in the homozygote shows a much more rapid progression and more severe end result than in the heterozygote.

We have described, in a general way, the course of the degenerative process, but it should be recognized that the process is rather variable, no two animals showing exactly the same appearance at the same age, and with some variation even between the two eyes of the same animal. Our observations, particularly those based on histological examinations, probably have not covered the spectrum of variability, and much remains to be learned by *in vivo* examination with ophthalmological equipment, more extensive histological studies using a variety of stains, immunological studies, and perhaps tissue culture. The occurrence of the mutant in an inbred strain is an advantage, particularly for immunological studies, since the effects of the mutant gene can be studied on an isogenic background.

The process appears to begin as a dissolution of lens fibres in the area surrounding the nucleus, and it is probably the altered refractive index in this region that allows one to see the transparent central body demarcated from the remainder of the transparent lens. As the region of degeneration spreads, its boundary advances peripherally, but not uniformly, giving the appearance of a 'pepper' shaped body with a region of transparent cortex around it. Further irregular progression of the boundary leads to the 'pseudopodial' stage. In certain regions the amorphous material reaches the capsule relatively early and can be seen as 'etched' streaks in the unfixed lens or dark areas in the fixed lens. As involvement of the cortex becomes more general, the shape of the central body becomes more spherical.

At least two other hereditary degenerative conditions of the lens are known in the mouse. The recessive mutant 'lens-rupture' (Fraser & Herer, 1950) bears no resemblance to Shrivelled. It is recessive, and characterized chiefly by extrusion of the nucleus at the posterior pole of the lens, a process never observed in Shrivelled animals. Conversely, the 'pepper' and 'pseudopodial' stages seen in Shrivelled lenses do not occur in the 'lens rupture' mutant. Preliminary observations suggest that the effects of the Shrivelled gene may suppress the expression of the *lr*/*lr* genotype more or less completely, possibly because the liquefied lens can no longer build up the necessary force to rupture the capsule.

On the other hand, the condition described as cataracta congenita subcapsularis by Paget & Baumgartner-Gamauf (1961) has many similarities to Shrivelled, but also some differences. The inheritance was dominant, but homozygotes did not show earlier onset or more rapid progression of the condition than did heterozygotes. Paget describes subcapsular liquefaction of the lens material, not involving the nucleus, which sounds very similar to the process observed in Shrivelled. In addition, however, Paget described passage of large amounts of liquid through the lens capsule with shrinkage of the lens, intense swelling of the retina, and persistence of the hyaloid artery, which we have not observed in Shrivelled. It is possible that the two mutations represent different alleles at the same locus, or that they are independent occurrences of the same mutation and that the differences in expression result from differences in the genetic background, or that they are mutations at different loci. The question cannot be solved until the two mutants have been intercrossed, but unfortunately this test cannot be made at present.

SUMMARY

Shrivelled, a new mutant in the house mouse, is characterized by degeneration of the fibres of the ocular lens, followed by the development of cataracts. The condition is caused by a mutant autosomal gene showing intermediate dominance.

REFERENCES

Fraser, F. C. & Herer, M. L. (1950). The inheritance and expression of the 'lens rupture' gene in the house mouse. *J. Hered.* **41**, 3–7.

Paget, O. E. & Baumgartner-Gamauf, M. (1961). Histologische Untersuchungen an einer dominant erblichen Form einer Cataract bei der Hausmaus. *Zool. Anz.* **166** (1–2), 55–69.