Zbl. Vet. Med. B, 24, 722—727 (1977)

ISSN 0514—7166/ASTM-Coden: ZVRBA2

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Influence of Feeding on Development of Nephritis and on Breeding Efficiency in Mice Infected Congenitally with Different Strains of LMC Virus

By

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With 2 tables

(Received for publication October 8, 1976)

During the last few years various commercial mouse feeds have been in use or on trial at this Institute. This offered an opportunity to study the influence of different diets on the development of glomerulonephritis, the dominant syndrome of "late onset disease" (1), and on the breeding capacity of congenital carriers of LCM virus. Results obtained will be described and commented on in the present communication.

Material and Methods

Mice and virus strains: Outbred white NMRI mice infected congenitally either with virus strain "W" or with "WCC" (4) originated from the persistently infected small breeding colonies established here in 1972.

Feeds: Commercial pellets produced by different firms were designated as "A" (adequate), "B" (inadequate) and "C" (poor, but better than B). The feed formula is known only for diet B. It comprises many different ingredients and appears to be a well balanced diet on paper.

Feed A was used here from 1972 to 1974. It was replaced by feed B late in 1974. Feed C was substituted for B several months later and has been in use since.

Diagnosis of nephritis: As in previous work, diagnoses could only be made macroscopically bas on the following criteria: illness; color changes of the kidneys (brown, yellowish, gray); increased kidney weight compared with normal mice of the same age and sex; enlargement of the suprarenal lymph nodes; in more acute cases serous exudation (pleural, peritoneal, subcutaneous) with exudate rich in lymphocytes, large mononuclears and many smudged cells; watery serum forming a heavy precipitate after inactiva-

tion at 58 °C for 30 min. and subsequent freezing and thawing; yield of exceptionally clear kidney extracts compared with the turbid and opalescing extracts obtained from normal looking kidneys. Moreover, diseased kidneys usually contain more specific complement-fixing (CF) antigen than those without macroscopic changes (4).

By macroscopic examination alone it is no doubt impossible to identify very mild cases of nephritis or early stages of the disease. Figures given in Table 1 therefore concern the more severe cases recognizable without histological examination.

Experiments and Results

Incidence of nephritis in congenital carriers receiving different feeds. Mice used in these experiments were observed for 8 months starting from birth. Cases of nephritis encountered during this period are listed in Table 1.

The data on mice given feed A were taken from a previous paper (4) and commented on there. Cases occurring after the 8th month of life in these 3 groups of animals (carriers of "W", carriers of "WCC" and normal mice) were omitted in the table.

Table 1

Incidence of nephritis in NMRI mice fed commercial pellets obtained from different sources

Feed	Mice		Cases of nephritis up to the age of 8 months		
	No.	status	with exudation	without exudation	total and percentage
A (good)	66	carriers of strain "W"	11	11	22 33.3 %
	16	carriers of strain "WCC"	2	3	5 31.2 %
	49	normal	0	1	1 2.0 %
B (poor)	39	carriers of "W"	0	0	0
	3	carriers of "WCC"	0	0	0
C (slightly better than B)	71	carriers of "W"	0	1	1 1.4 %
	64	carriers of "WCC"	4	6	10 15.6 %

In 42 mice receiving feed B nephritis was not seen. The diet was able to maintain the animals in good health although many of them appeared rather thin. No fat deposits were seen at autopsy. It is unfortunate that this batch, originally intended for collection of serum and organ materials at different age levels, contained only 3 carriers of "WCC". These were scarce at the time of assembly of the group.

A surprising result was obtained from mice maintained on diet C. Whereas only 1 mouse, 207 days old, among 71 carriers of strain "W" showed typical nephritis, 10 cases of the disease were found among 64 animals infected with "WCC". The affected mice were 144—245 days old at the time of autopsy.

Breeding efficiency of mice kept on different diets. Very good breeding results were obtained with NMRI mice as long as feed A was available. First

litters from uninfected females averaged 11.3 baby mice and those from carriers of "W" or "WCC" 10.5 and 9.4 sucklings, respectively (4). There were no sterile animals, no abortions and cannibalism was at a minimum.

The situation changed drastically when feed B was used. Its harmful effect on the breeding capacity of females was first noticed with gray mice of breed CBA/J (inbred animals carrying strain "W"). They failed to produce a single litter in several months. The result obtained with NMRI mice were only slightly better as can be seen in Table 2. Some females did not conceive, others aborted at different stages of pregnancy or had stillbirths, and still others ate up their litters entirely or in part. In many cases the basic reason for this cannibalism seemed to be the mother's inability to lactate.

A changeover to feed C brought some improvement and enabled us to maintain the three colonies of congenital carriers. The effect was most marked with inbred CBA/J mice which promptly regained their former breeding efficiency. Results obtained with NMRI mice from successive generations (see figures in column 1) are recorded in Table 2. The females of generation 1 remained with the males up to the age of 8 months. Those of generation 2 were used for other purposes after the second or third pregnancy. The breeding test with generation 3 is still in progress. An upward trend is recognizable with carriers of strain "W" but still missing in those of "WCC".

Table 2
Breeding results

Feed	Breeders: virus carried	Breeders: No. of ♀	Breeders: sterile	Mean numbers of live baby mice from successive pregnancies — Pregnancy No. 1	2	3	4	5	6
A	"W" / "WCC"	see ref. (3), tables 1 and 2							
B	"W"	14	3	11* / 1.8	7 / 4.3	6 / 2.8	2 / 2		
	"WCC"	8	2	6 / 4.3	4 / 0	2 / 2	2 / 0		
C 1	"W"	23	1	22 / 3.2	18 / 4.5	17 / 5.2	16 / 4.9	11 / 5.9	4 / 2.5
	"WCC"	16	2	14 / 4.9	12 / 6.4	7 / 5.1	3 / 4.3	2 / 3.0	
C 2	"W"	15	2	13 / 2.3	9 / 2.9	2 / 8.5			
	"WCC"	9	2	7 / 3.9	4 / 3.5	1 / 0			
C 3	"W"	6	0	6 / 3.5	6 / 6.7	4 / 7.2			
	"WCC"	7	0	7 / 4.1	2 / 3.0				

* first litters of 11 females averaged 1.8 baby mice born alive and nursed by the mothers

Discussion

Before incriminating faulty nutrition as the cause of the numerical decrease of nephritis cases and the reduced breeding capacity in persistently in-

fected NMRI mice it is necessary to exclude another factor which might theoretically be responsible for the changes observed, namely, prolonged natural (vertical) passage. Strong evidence against this possibility comes from the famous HAAS colony in which chronic LCM infection has persisted for more than 30 years. A high incidence of glomerulonephritis and good breeding results have recently been reported for these mice (3). The return of our persistently infected CBA/J mice to normal breeding after a change of feed (B to C) should also be considered in this connection even though conclusions concerning the frequency of nephritis are not possible in this case since the nephritis rate is very low in CBA/J carriers even under optimal nutritional conditions (5).

From the results obtained here with NMRI mice and the behavior of HAAS mice, which undoubtedly are well nourished (3), it may be concluded that a nutritional deficiency was responsible both for the reduced incidence of nephritis and the poor breeding results in our carriers of strain "W".

On the other hand, 15.6 % of the mice persistently infected with "WCC" and kept on the same diet developed nephritis in spite of the fact that their breeding record was even worse than that of females carrying strain "W". This shows that the parallelism between frequency of nephritis and breeding efficiency observable in carriers of strain "W" was missing in mice infected with "WCC". The different behavior of "W" and "WCC"-infected animals in this respect points to a direct pathogenic effect of the virus becoming manifest when viral concentration has reached a critical high level in the kidneys. In previous experiments (4, Fig. 1 and 2) renal antigen titers were strable by indirect CF did not differ significantly in carriers of "W" and on the average higher in carriers of "WCC" than in those of strain "W" and nephritic kidneys as a rule contained more viral antigen than macroscopically normal ones. It seems possible that in mice with nutritional deficiency "WCC" virus can reach this critical concentration more readily than strain "W".

The percentages of animals which developed specific antibodies demon- on the average higher in carriers of "WCC" than in those of strain "W" and "WCC". It is therefore unlikely that intensified antibody production in "WCC" mice led to increased formation of antigen-antibody complexes believed to have a harmful effect upon the glomeruli of the kidneys (2).

For technical reasons we have not been able to search for the essential ingredient(s) missing or insufficient in amount in the more or less inadequate diets B and C. We suspect that a quantitative or qualitative protein deficiency is responsible.

Summary

In NMRI mice infected congenitally with strain "W" of LCM virus a changeover to obviously deficient commercial diets resulted in a marked reduction of both the incidence of glomerulonephritis and the breeding capacity in such animals.

On the other hand, congenital carriers of LCM strain "WCC" fed in the same way developed severe nephritis 10 times more frequently than their "W"-infected counterparts even though their breeding efficiency was slightly lower. This shows that a parallelism between incidence of nephritis and breeding efficiency, noticeable in carriers of strain "W", was missing in those of "WCC".

The bearing which these results may have on the pathogenesis of late-onset nephritis is discussed.

Acknowledgement

We thank Prof. Dr. Dr. h. c. ANTON MAYR for providing laboratory facilities and materials for this study.

Zusammenfassung

Einfluß der Fütterung auf die Entstehung von Nephritis und die Zuchtfähigkeit bei congenital mit verschiedenen LCM-Virusstämmen infizierten Mäusen

Bei congenital mit dem Stamm „W" des LCM-Virus infizierten NMRI-Mäusen bewirkte der Übergang zu offensichtlich mangelhaftem, handelsüblichem Mäusefutter einen starken Rückgang sowohl der Glomerulonephritisfälle als auch der Zuchtfähigkeit dieser Tiere.

Andererseits erkrankten congenital infizierte Virusträger des Stammes „WCC" bei gleicher Ernährung 10mal häufiger an schwerer Nephritis als die erstgenannten Mäuse, obwohl ihre Vermehrungsfähigkeit noch stärker beeinträchtigt war. Ein Parallelismus zwischen Nephritishäufigkeit und Zuchtfähigkeit wie bei den mit dem Stamm „W" infizierten Tieren fehlte also bei „WCC"-Virusträgern.

Mögliche Zusammenhänge zwischen diesen Befunden und der Pathogenese der Spätnephritis werden besprochen.

Résumé

Influence de l'alimentation sur l'apparition de néphrite et sur la reproduction chez des souris infectées congénitalement par différentes souches virales LCM

Chez des souris NMRI infectées congénitalement par une souche «W» du virus LCM, le passage à une alimentation manifestement carencée provoqua un fort recul des cas de glomérulonéphrite et de la capacité de reproduction de ces animaux.

Les porteurs de virus de la souche «WCC» infectés congénitalement présentèrent avec la même alimentation dix fois plus de néphrites graves et une atteinte encore plus grave sur leur capacité de reproduction. Il n'y a pas eu de parallélisme entre la fréquence des néphrites et la capacité de reproduction chez les porteurs du virus «WCC» comme ce fut le cas chez les animaux infectés avec la souche «W».

On discute les rapports possibles entre ces résultats et la pathogénèse de la néphrite retardée.

Resumen

Influjo de la alimentación sobre la patogenia de la nefritis y la aptitud zootécnica de ratones infectados congénitamente con diversas estirpes virales de LCM

En ratones NMRI, infectados congénitamente con la estirpe «W» del virus LCM, ocasionaba la transición a pienso comercial para ratones, deficiente evidente, un retroceso intenso tanto de los casos de glomerulonefritis como de la aptitud zootécnica de estos animales.

Por otro lado, enfermaron los portadores de virus de la estirpe «WCC», infectados por vía congénita, con la misma alimentación, de nefritis con una

frecuencia 10 veces mayor que los ratones citados en primer lugar, aunque su capacidad reproductora se hallaba amenguada aún mucho más. Por tanto, en los portadores de virus «WCC» faltaba un paralelismo entre la frecuencia de nefritis y la aptitud zootécnica como en los animales infectados con la cepa «W».

Se discuten las conexiones posibles entre estos hallazgos y la patogenia de la nefritis tardía.

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