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STUDIES ON ECHINOCOCCOSIS XIV
FURTHER OBSERVATIONS ON THE DIFFERENCE OF SUSCEPTIBILITY TO ECHINOCoccus MULTilocULARIS AMONG UNIFORM STRAINS OF THE MOUSE

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(Received for publication, March 7, 1963)

The authors (1958) published their results of investigations on experimental primary infection of Echinococcus multilocularis in various rodent animals among which 10 uniform strains of the mouse were included. They studied the difference of susceptibility by species and strains of the animals and clarified the following facts. The lowest susceptibility, 8%, was observed in dd strain and the highest, 100%, in 3 strains such as AKR for one. The difference was found not only in the infection rate but also in the type of the lesion; the authors differentiated two types of lesion which were induced by combination of morphology and developmental speed of the echinococcus larva and by host tissue reactions. As for the types of the multilocular echinococcosis, one of the authors discussed the matter later in detail from the viewpoint of histo- and pathogenesis (OHBAYASHI, 1960).

Other than the authors' foregoing reports, results pertaining to the susceptibility in various animals have been published by VOGEL (1955, 1955), RAUSCH and SCHILLER (1956) and SADUN et al. (1957). Recently, WEBSTER and CAMERON (1961) also made experimental investigations about the same problem.

The authors have carried out a series of experiments on the susceptibility and they, in the present paper, wish to publish the results obtained from an experiment on five strains of the mouse.

MATERIALS AND METHODS

Five mouse strains were used; 8 animals of Fm strain, 18 of NC, 22 of SM, 21 of gpc and 43 of KK. The animals were obtained from the breeding stocks of the Institute for Infectious Diseases, University of Tokyo. The mice were given inoculation of physiological
saline solution with the eggs by a syringe with cannula. Oral infections were conducted: the inocula which contained eggs of the Alaskan strain of *Echinococcus multilocularis* were obtained from the feces or from adult tapeworms originated in experimentally infected dogs dissected more than 5 weeks after oral infection of the hepatic multilocular echinococcus. The number of eggs inoculated was about 100–150 in the fecal material and about 200–300 in the cestode material.

The mice inoculated were sacrificed at desired intervals and examined macro- and microscopically by routine methods.

**RESULTS**

The echinococcus foci were limited to the liver in all cases.

**Strain fm** 2♂♂, 6♀♀ (2; 1/X'59–18/I'60; 4 1/2). [Data show, in order, the number of animals and their sexes (age in months when inoculated; days inoculated–killed; duration after the inoculation in months)]. Microscopicals: Echinococcus foci in all cases; the foci circumscribed and small-cystic multilocular form in general; one case, however, with widely spread foci. Microscopicals: Severe granulation tissue reaction; tendency of calcification in contact with the echinococcus cyst; scolex formation in only one case.

**Strain NC** 18♂♂ (6; 10/I'60–10/XII'60; 3). Macro.: Positive in 7 cases; small-cystic multilocular in general; rarely large cysts. Micro.: Remarkable formation of granulation tissue around the cysts; rare formation of the initial brood capsule in one case.

**Strain SM**  a) 18♂♂ (4; 1/X'59–23/X'59, 29/X'59, 18/I'60; 1 4/5, 2, 4 1/2). Macro.: Positive in all cases; the foci circumscribed and small-cystic multilocular in general. Micro.: Remarkable proliferation of granulation tissue; tendency of necrosis in contact with the cyst wall.

b) 4♂♂ (4; 29/I'60–14/X'I'61; 12). Macro.: Two positive cases. Micro.: Fully developed with remarkable formation of scolices; the echinococcus itself sometimes with regressive changes.

**Strain gcc**  a) 14♂♂ (9; 1/X'59–18/I'60; 4 1/2), b) 7♂♂ (2; 1/X'59–21/X'59, 19/X'59, 18/I'60; 1 2/3, 2 2/3, 4 1/2). Macro.: 15 positive cases out of 21. Micro.: Formation of initial scolices in one case of 4 months and a half after the infection; in other cases the echinococcus surrounded by marked reaction of granulation tissue; frequently regressive findings in granulation tissue.

**Strain KK**  a) 17♂♂ (4; 10/I'60–10/XII'60; 3). Macro.: All cases positive; the foci widely spread and the liver swollen; many large cysts. Micro.: Scolex formation in 10 cases; remarkable completed scolices.

b) 19♀♀ (4; 10/I'60–10/XII'60; 3). Macro.: All cases positive; distribution of the foci not so good as in group a. Micro.: Some large cysts, but small-cystic multilocular echinococcus in general. Micro.: Active proliferation of granulation tissue; formation of initial scolices and brood capsules in one case.

c) 1♀ (5; 10/I'60–24/I'61; 4 1/2). Macro.: Positive. Micro.: The echinococcus with advanced scolex formation.

d) 6♀♀ (7; 5/I'60–14/X'I'61; 14). Macro.: Positive in 2 cases; fully developed larval echinococcus with active formation of scolies; regressive changes in the echinococcus in some
DISCUSSION

The experimental animals, in the present work, were mostly dissected 3~4 months after the inoculation for the purpose of examining and classifying the echinococcal lesions. Duration of 3~4 months is considered adequate for comparison of present findings with those of preceding report (1958). The authors, in the foregoing report, classified the lesions of experimental animals into 2 types as follows. Type 1 is characterized by rapid development of the echinococcus in which many large cysts are noted, the scolex formation in earlier period such as 1.5~2 months after the infection and slight host tissue reactions. Contrary to the above, type 2 manifests slow development, small-cystic multilocular structure, delayed scolex formation such as 5~7 months and severe host tissue reactions. In this classification, it is easily understood that the suitability of animals as the intermediate host has a close relation to appearance of the difference, in other words, the rapidity of development plays an important role. After long post-infective period, the echinococcus of type 2 also progresses until the completion of its structure takes place and the difference between types 1 and 2 becomes indistinct superficially. In reality, however, the difference can still be noted in a portion of the multilocular vesiculation, characteristic of tissue reaction and other findings. Length of life of host animals after the inoculation naturally shows notable difference according to the type. At any rate, it can be concluded that there exists remarkable difference of susceptibility among species and strains of host animals.

WEBSTER and CAMERON (1961) recorded experimental infections in rodents with *Echinococcus multilocularis sibiricensis* (identical to the present authors' strain), *E. granulosus canadensis*, and *E. granulosus* (Lebanon origin). They discussed host-parasite relationship and considered the strain of host animals as a factor to

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<th>STRAINS OF MICE</th>
<th>NO. OF CASES OF WHICH RESULTS WERE</th>
<th>SUSCEPTIBILITIES (%)</th>
<th>TYPES*</th>
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<tbody>
<tr>
<td>fm</td>
<td>8 0</td>
<td>100</td>
<td>2</td>
</tr>
<tr>
<td>NC</td>
<td>7 11</td>
<td>39</td>
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</tr>
<tr>
<td>SM</td>
<td>18 4</td>
<td>82</td>
<td>2</td>
</tr>
<tr>
<td>gpc</td>
<td>15 6</td>
<td>71</td>
<td>2</td>
</tr>
<tr>
<td>KK 5</td>
<td>17 0</td>
<td>100</td>
<td>1</td>
</tr>
<tr>
<td>9</td>
<td>22 4</td>
<td>85</td>
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*: Classification of the multilocular echinococcosis by the authors (1958).
influence susceptibility. As for the mouse, they examined the susceptibility both of strains DBA/1J and C57BL/6J to *E. multilocularis sibiricensis*, with results obtained differing from those of the present authors. However, there is full agreement, the authors consider, that WEBSTER and CAMERON attach importance to the genetic constitution of experimental animals.

Results presently obtained are summarized in table 1. Various rates of susceptibility are noted in the same way as in the preceding report. Among those strains, strain KK becomes a particular subject of discussion; in male group of strain KK, the echinococcus foci belong to type 1 and in the female, to type 2. Infection rate of female group is also inferior to that of male group.

There have been published many reports in connection with age and sex resistance of host animals to infection. As to the sex resistance to nematode infection, published results about *Ascaridia galli, Aspiculuris tetraptera, Syngamus trachea* and *Nippostrongylus muris* are available. Especially in the experiment by HALEY (1958) on this resistance of hamsters to *Nippostrongylus muris*, intense resistance of female hamsters was shown. Among cestode parasites, *Hymenolepis diminuta* and *Cysticercus fasciolaris* have been frequently dealt with by many authors and the sex resistance in female host has been also clarified. In almost all these reports, the number of parasites is the ground of argument. The multilocular echinococcus in male animals of strain KK in the present work developed as the echinococcus in a suitable host, but in the female, it developed as in an unsuitable host. SADUN (1951) investigated the effect of homologous gonadal hormones in experimental *Ascaridia galli* infection in chickens; he found an effect upon the growth of the parasite. He clarified the fact that injections of a-estradiol benzoate seemed to cause a temporary retardation, and that of testosterone propionate a temporary acceleration. Comparing theirs with SADUN's results, the authors would like to explain the findings of strain KK as a phenomenon of the same category.

**SUMMARY AND CONCLUSION**

Primary infections of *Echinococcus multilocularis* were examined experimentally on 5 mouse strains fm, NC, SM, gpc and KK; the susceptibility to the parasite was investigated. As to infection rates, strains fm and KK showed high rates, NC low, and SM and gpc intermediate. Strains fm, NC, SM and gpc manifested type 2 lesions of echinococcosis. The lesions in both sexes of strain KK were classified into different types: male mice were affected by type 1 lesion and female type 2. It can be concluded that strain KK shows sex resistance in female cases to the multilocular echinococcus infection.
REferences

2) HALLEY, A. J. (1958): Ibid., 7, 338
6) SADUN, E. H. (1951): Exp. Parasit., 1, 70

Explaination of Plate

Fig. 1. Strain fm, 4 l/2 months after the infection; multilocular echinococcus with tissue reaction. Hematoxylin-eosin (H.-E.), × 50
Fig. 2. NC, 3 months; severe tissue reaction. H.-E., × 50
Fig. 3. SM, 3 months; multilocular echinococcus without brood capsule formation. H.-E., × 50
Fig. 4. gpc, 4 l/2 months; typical multilocular echinococcus. H.-E., × 50
Fig. 5. KK, ♂, 3 months; liver lesions. × 2/5
Fig. 6. KK, ♀, 3 months; liver lesions. × 2/5
Fig. 7. KK, ♂, 3 months; scolex formation. H.-E., × 50
Fig. 8. KK, ♀, 3 months; initial formation of brood capsule. H.-E., × 50