by way of the local peripheral nerves). Among 21-day-old mice, however, 70 to 80% develop flaccid paralysis, 10 to 20% exhibit only signs of encephalitis, and the few remaining ones show no evidence of CNS disease. About half the number of 7- to 10-day-old guinea pigs injected into the leg muscles with the mouse-passage strain of W.E.E. exhibit only signs of encephalitis; an occasional young animal shows no nervous signs at all but most of the others reveal flaccid paralysis of the posterior extremities (first on the inoculated side) with pathological evidence of spread only along the local peripheral nerves and none of the diffuse vascular and perivascular lesions in the neopallial cortex. Since the capacity to produce such a high incidence of flaccid paralysis following injection into the leg muscles has not been noted previously in work on W.E.E., it is not improbable that it may be related to the number of mouse passages our strain of virus has undergone.

9949 P

Age of Host and Capacity of Equine Encephalomyelitic Viruses to Invade the CNS.

ALBERT B. SABIN AND PETER K. OLITSKY.
From the Laboratories of the Rockefeller Institute for Medical Research, New York.

Studies on the nature of various types of natural resistance to viruses affecting the central nervous system (CNS) have already disclosed that as mice and guinea pigs grow older there appear certain localized barriers in the peripheral and central nervous systems which, because of the nature of neural progression of peripherally injected vesicular-stomatitis virus, are capable of preventing it from invading the CNS or from spreading beyond a certain point, depending upon the route of inoculation.[1] Still another type of change that occurs with age was revealed in the course of investigations on the manner in which the viruses of equine encephalomyelitis (E.E.) invade the CNS after inoculation into the muscles of an extremity. This change which appears to involve the blood vessels, which the E.E. viruses must traverse or grow through in order to invade the CNS,[2, 3, 4] is the subject of the present report.

[1] Sabin, A. B., and Olitsky, P. K., *J. Exp. Med.*, 1937, **66**, 15, 35; 1938, **67**, 201, 229.

[2] Sabin, A. B., and Olitsky, P. K., *Am. J. Path.*, 1937, **13**, 615.

[3] Sabin, A. B., and Olitsky, P. K., PROC. SOC. EXP. BIOL. AND MED., this issue.

[4] Hurst, E. W., *J. Path. and Bact.*, 1936, **42**, 271.

With the Eastern strain of the mouse-passage virus 15-day-old mice are 100% susceptible (intramuscular or intraäbdominal inoculations) while at one month of age 40 to 50% are already resistant and beyond the age of 3 months this resistance rises to 95%. Studies on the nature of this resistance revealed that while the virus invades (and persists or multiplies in) the circulating blood as in young mice, it apparently cannot get across the vessels, which for this infective agent seems to be in most instances a necessary preliminary step to involvement of the nervous system. It has already been shown[5] that young and old mice are practically equally susceptible to intracerebral and intranasal administration of the virus further suggesting that some change in the blood vessels is probably responsible for the resistance of the older animals. It is of interest to note that, as with vesicular-stomatitis virus,[1] the majority of old mice which are resistant to intramuscular inoculation succumb when the E.E.E. virus is injected directly into the sciatic nerve. In guinea pigs inoculated intramuscularly with 10^8 or 10^9 mouse-cerebral lethal doses (M.C.L.D.) no difference was observed with age, while with 10^7 or fewer M.C.L.D., old guinea pigs (800-1000 g) resisted in simultaneous tests in which 7- to 10-day-old animals (60-100 g) succumbed; one of the old guinea pigs injected with the smaller dose developed flaccid paralysis of the posterior extremities and exhibited lesions indicative of invasion along the nerves supplying the inoculated muscle, but not the diffuse and widely scattered vascular and perivascular changes ordinarily encountered in this species.[3]

With the Western strain of the mouse-passage virus (W.E.E.) the first evidence that age has produced some change in the animal is found in the fact already noted[3] that the majority (80 to 90%) of 15-day-old mice develop signs of encephalitis and only a few animals signs of flaccid paralysis after inoculation of the virus into one of the posterior extremities, while at 21 days of age the ratio is reversed, the majority developing flaccid paralysis and some (6 of 30) resisting altogether. At 1 month of age* and beyond, 90% or more of mice are resistant to intramuscular inoculation, (of 36 mice older than 6 months only one succumbed) although direct injection into the sciatic nerve supplying these muscles again results in the paralytic disease in a large proportion of the animals. There are thus at least 3 phases during the maturation of the mouse at

[5] Olitsky, P. K., Sabin, A. B., and Cox, H. R., *J. Exp. Med.*, 1936, **64**, 723.

* Resistance to intramuscular injection of vesicular stomatitis virus also appears at this age and is just as marked.

which the neuroinvasiveness of W.E.E. virus is differently affected: first, at the very early age of 15 days when certain vessels still permit the virus to traverse and spread in the CNS by some definite pathway;[3] second, at the age of 21 days when the virus can no longer do this in the majority of mice but now progresses along the nerves supplying the inoculated muscle; and third, between the 21st and 30th days of life when the appearance of some change in the muscle or specialized nerve endings in the great majority of mice (it is important to remember the few exceptions) now prevents invasion of the CNS altogether. Intramuscular injections of as much as 10^8 M.C.L.D. of the mouse-passage W.E.E. virus finds practically all old guinea pigs (800-1000 g) resistant with only an occasional one exhibiting flaccid paralysis.

Conclusions. Studies of the conditions which permit or prevent the viruses of vesicular stomatitis and equine encephalomyelitis to invade the CNS of mice and guinea pigs have thus far disclosed that depending upon the age or species of the host there may be changes or variations in localized sites, *e.g.*, in terminal, specialized nerve endings, in special areas of the CNS, blood vessels, etc., which can prevent the virus from invading or progressing in the CNS—changes in different tissues functioning as barriers for different viruses depending upon the mode of CNS invasion utilized by the virus.

9950 P

Inhibition of Estrogenic Effects on the Skeleton by Testosterone Injections.*

W. U. GARDNER AND C. A. PFEIFFER.

From the Department of Anatomy, Yale University School of Medicine.

Hypercalcification of the skeleton involving a partial or complete obliteration of the marrow cavities occurs in immature rats and chickens[1] and in mice[2] following extended treatment with large amounts of estrogens. Simultaneously with this, the pubic and ischial bones are reabsorbed at the symphysis and the former carti-

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[1] Zondek, B., *Folia Clin. Orient.*, 1937, **1**, 1.