suspending media were saline, 4% phosphorylated hesperidin solution and a water-in-oil emulsion. In each case the amount injected consisted of one cc of material containing 500 million organisms/cc. The water-in-oil emulsion was composed of 2 parts saline suspension of organisms, 2 parts paraffin oil and one part G 1096 Atlas emulsifier. A booster injection was given at the end of the 3rd week. Blood samples were collected each week and the antibody titration carried out by means of the standard agglutination test.

The results are pictured graphically in Fig. 1 which shows the unusual effectiveness of phosphorylated hesperidin as an adjuvant. Figures are averages for the 6 rabbits in each group. In view of the current interest in gamma globulin for passive immunization against poliomyelitis, and the relative shortness of the period of effectiveness, it is intended to determine if the use of phosphorylated hesperidin might extend the period of protection now offered by gamma globulin.

*Summary.* An adjuvant action of phosphorylated hesperidin in the induction of the antibody response to typhoid vaccine has been demonstrated.

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## Influence of Low Tryptophan Diets Containing 6-Methyltryptophan on Oral Infection with Poliomyelitis Virus.* (20627)

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A study of the relation of the nutritional state of the host to susceptibility to Lansing (Type II) poliomyelitis in mice has established the fact that the development of Lansing infections is altered in animals suffering from a number of nutritional deficiencies (1). Tryptophan deficiency is particularly effective in modifying poliomyelitis in mice (2,3) and mice may be partially protected from Lansing virus by feeding the tryptophan analogue, 6-methyltryptophan(4). Preliminary experiments indicating that low tryptophan diets containing 6-methyltryptophan also exert some protective action against oral infection with poliomyelitis virus in monkeys are described here.

*Toxicity of 6-methyltryptophan.* Our studies in mice(4,5) had shown that the greatest protection against Lansing infections was observed in the animals fed diets containing a small amount of tryptophan (0.05%) and 10 times that amount of 6-methyltryptophan. The tolerance of 1.2 to 2.2 kg Philippine cynomolgus monkeys‡ for 6-methyltryptophan§ (6-MT) was approximated by feeding 6-MT in a diet consisting of: Corn grits,‖ 89%; corn oil¶ (fortified with oleum percomorphum** 25 ml/liter), 5.5%; Salts "IV" of Phillips and Hart(6), 4%; 1-lysine HCl, 1.0%; and DL methionine, 0.5%. Water soluble vitamins

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‡ Obtained from Okatie Farms.

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‖ Quaker "Hominy Grits".

¶ Mazola.

** Mead Johnson and Co.

TABLE I. Summary of Experiments Showing Influence of Diets Containing 6-Methyltryptophan (6-MT) on Susceptibility of Cynomolgus Monkeys to Oral Infection with Poliomyelitis.

| Exp. 1: 2% 6-MT, 0.05% tryptophan in corn grit basal | | | | | | Exp. 2: 2% 6-MT in corn grit basal | | | | | | Exp. 3: 2% 6-MT in corn grit basal | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 6-MT | | | M-3 control | | | 6-MT | | | M-3 control | | | 6-MT | | | M-3 control | | |
| No.* | F† | P‡ | No. | F | P | No. | F | P | No. | F | P | No. | F | P | No. | F | P |
| 1 | 0 | 8 | 17 | 5 | 6 | 31 | 5 | 7 | 37 | 6 | 7‖ | 57 | 7 | 0¶ | 67 | 3 | 10 |
| 6 | 0 | 8 | 24 | 4 | 7 | 32 | 3 | 0 | 43 | 6 | 8 | 51 | 6 | 0 | 62 | 9 | 12 |
| 11 | 0 | 8 | 16 | 5 | 8 | 25 | 0 | 0 | 36 | 5 | 8 | 53 | 6 | 0 | 64 | 12 | 13 |
| 8 | 7 | 9 | 21 | 7 | 8 | 26 | 0 | 0 | 41 | 9 | 10 | 49 | 0 | 0 | 63 | 3 | 30 |
| 15 | 9 | 9 | 20 | 8 | 8 | 27 | 0 | 0 | 38 | 6 | 11 | 50 | 0 | 0 | 59 | 3 | 0 |
| 9 | 9 | 10 | 18 | 7 | 9 | 28 | 0 | 0 | 42 | 7 | 12 | 52 | 0 | 0 | 68 | 4 | 0 |
| 13 | 0 | 10 | 22 | 8 | 9 | 29 | 0 | 0 | 35 | 0 | 14 | 54 | 0 | 0 | 66 | 6 | 0 |
| 3 | 0 | 0§ | 23 | 8 | 9 | 30 | 0 | 0 | 44 | 0 | 19 | 55 | 0 | 0 | 61 | 0 | 0 |
| 4 | 0 | 0§ | 19 | 8 | 11 | 33 | 0 | 0 | 39 | 0 | 0 | 56 | 0 | 0 | 65 | 0 | 0 |
| | | | | | | 34 | 0 | 0 | 40 | 0 | 0 | 58 | 0 | 0 | | | |
| Total paralysis | | 7 | | | 9 | | | 1 | | | 8 | | | 0 | | | 4 |

* Monkey No.

† F = Days after first virus feeding on which fever of 103°F or higher developed.

‡ P = " " " " " when paralysis appeared.

§ Monkeys No. 3 and No. 4 resisted intracerebral challenge with 500 $LD_{50}$ Wisconsin '45 virus 6 weeks after oral inoculation. All other surviving animals developed typical poliomyelitis following intracerebral challenge.

‖ Monkey No. 37 was found dead on the 7th day. Examination of the brain and cord revealed typical histopathology of severe poliomyelitis.

¶ Monkey No. 57 was slow, dull and ataxic 9 days after inoculation with virus but recovered without developing paralysis.

were provided as a supplement in the drinking water each morning(7). Control animals were fed our basal ration (M-3) containing 18% casein(7). The addition of 2% 6-MT to the corn grit basal, either with 0.05-0.1% supplementary tryptophan or without had no consistent adverse influence, as compared to the corn grit basal alone. Some animals lost weight rapidly on these rations, but so also did animals on the corn grit basal without 6-MT; and occasional animals failed to thrive even on 18% casein.

Subsequent nutritional observations were limited to those animals which also received virus (Exp. 1, 2 and 3, Table I) and definite conclusions as to the chronic toxicity of 6-MT cannot be drawn. It would appear that, as has been shown to be true in the mouse(5), 6-MT is not a highly active nutritional antagonist for tryptophan in the monkey. Several of the monkeys in Exp. 2 and 3 (Table I) developed anorexia during the last days of the experiment and it is thought that this may have been a manifestation of chronic tryptophan deficiency. On the other hand, it is probable that some animals would tolerate 2% 6-MT in the corn grit basal for a longer time and that concentrations considerably greater than 2% could be fed for shorter periods. Although a general tendency to diarrhea was observed among all of the monkeys including those receiving 18% casein, none developed the severe dysentery characteristic of deficiencies of folic acid and other water soluble vitamins.

*Influence of 6-MT on oral infection.* Philippine cynomolgus monkeys weighing from 1,200 to 2,200 g and the Wisconsin '45 strain of Type I poliomyelitis virus in the form of a pool of 10% monkey brain-cord suspension, prepared and stored in 50 ml aliquots at –40°C, were employed. The monkeys were inoculated by offering them 20 ml of virus suspension, sweetened with sucrose, in their drinking cups. Water was withheld on the day or days virus was given until the virus suspension was consumed. Ordinarily, the animals drank the virus suspension immediately. The indicated amounts of 6-MT and tryptophan were added to the dry corn grit basal, moistened with water and cooked in the autoclave for 30 minutes at two pounds pressure. The resulting ration was granular but soft and was much

more palatable to the monkeys than was a dry ration. The ration was offered in small amounts, about 50 g, 3 times daily. Control animals received the 18% casein basal(7).

In the first experiment, a group of 9 monkeys was placed on the basal ration containing 2% 6-MT and 0.05% dl-tryptophan for 7 days; and on the 8th, 9th, and 10th days this group and a control group of 9 monkeys were fed a total of 120 ml of 10% virus suspension (about 1,200,000 intracerebral $LD_{50}$) in 6 doses of 20 ml each. Temperatures were recorded daily and the animals were observed for signs of disease twice daily. Three weeks after the first virus feeding the surviving animals were placed on the 18% casein basal diet. The results are recorded in Table I.

Exp. 2 and 3 (Table I) were similar except that 1) the 2% 6-MT ration was not supplemented with tryptophan and 2) the monkeys received two feedings of 20 ml of a 1:400 dilution (approximately 10,000 intracerebral $LD_{50}$) from the same pool of Wisconsin '45 virus on the eighth day of the experiment. The surviving animals of all 3 experiments were challenged intracerebrally with 500 $LD_{50}$ Wisconsin '45 virus 6-9 weeks after their oral exposure.

In Exp. 1, there was a suggestion of protection against the virus in the 6-MT group, *i.e.*, incubation periods were longer, febrile prodromata less pronounced and two animals escaped paralytic infection. These 2 animals, No. 3 and 4, apparently did experience an inapparent infection for they resisted a large intracerebral challenge 9 weeks later. In the second and third experiments, the smaller inoculum resulted in a lower incidence of paralytic infection in the controls, 8/10 and 4/9 respectively, while only one of the 20 animals receiving 6-MT became paralyzed. The surviving animals in the latter experiment succumbed to intracerebral challenge.

*Summary.* These experiments are obviously of a preliminary nature but they do show that the susceptibility of a primate to poliomyelitis via a natural portal of entry may be altered and they suggest that this approach to the modification of susceptibility to poliomyelitis should be explored further.

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## Effect of Adrenocortical Steroids on Duration of Pentothal Anesthesia in Adrenalectomized Mice. (20628)

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It is well known that the adrenalectomized animal becomes less resistant to various types of damaging agents and more sensitive to various drugs, including anesthetics. And recently, much has been published on the central action of adrenocortical steroids. It is supposed from these that the adrenal gland may play an important role in anesthesia. The present experiment was performed to confirm this.

*Materials and methods.* The animals used in this study were 60 female albino mice weighing about 20 g, divided into 6 groups of 10 mice each, as shown in Table I. The