

in the form of boiled polished rice to which was added an additional amount of dried brewer's yeast. Several other puppies were fed this completed milk for shorter periods with satisfactory results.

Summary. A synthetic milk has been produced that has been found satisfactory for vitamin A studies on young puppies. This milk could also serve for studies on vitamins B, G, and D.

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Preliminary Observations on the Frei Test in Lymphogranuloma Inguinale.

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Since the Frei test is considered to be allergic in nature a passive transfer in the sense of Prausnitz and Küstner was attempted. Five non-tuberculous individuals who had reacted negatively to Frei antigens and one Rhesus monkey were intradermally inoculated in different skin areas of the back with 0.1 cc. Frei antigen, 0.1 cc. inactivated normal human serum, 0.1 cc. sera of typical cases of Lymphogranuloma inguinale and 0.1 cc. mixtures of normal human serum and Frei antigens. Twenty-four hours later 0.1 cc. of a potent Frei antigen was injected into the same areas but with negative results. This was repeated in 3 patients at the end of 1 and of 2 months, again with negative findings.

The Frei test as now used is often difficult to evaluate. It is evident from the reactions that the antigen far exceeds the necessary dose for a more specific and less potent reaction and that the antigens may also contain non-specific factors.

Samples of Lymphogranuloma inguinale pus were dried *in vacuo* above anhydrous CaCl_2 , ground to a fine powder and weighed. It was found that 0.1 gm. suspended in 10 cc. of physiologic salt solution (1:100) and heated at 60°C. gives an excellent antigenic emulsion. A positive reaction was secured in one case in a dilution as high as 1:20,000.

However, the grading off usually occurred between 1:5,000-1:10,000. Therefore, the test can be performed with much more dilute antigens than are at present in use. High speed centrifugations of the antigens yielded non-reactive supernatant liquids. Berkefeld filtrates also gave negative results.

Acidification or alkalization of the dried antigen with N/100 HCl or N/100 NaOH and neutralization prior to injection did not destroy the reacting factors. The residual lipoids from an alcohol ether extraction (3:1) were non-reactive. The protein fraction, however, gave an active antigen.

Sera from cases of Lymphogranuloma inguinale did not neutralize the Frei antigens. Recently Reiss¹ concluded that the serum of an early case of Lymphogranuloma inguinale possesses antigenic properties. However, he did not use a normal serum control. We repeated this work in a series of 20 individuals including 6 Frei positive cases with negative results in all cases of Lymphogranuloma inguinale. Our observation is in accord with the findings of Levaditi and Reinié² that the plasma of infected monkeys does not carry the virus but that the virus may occasionally occur in white blood corpuscles and lymph.

Fresh pus from typical cases yielding potent Frei antigens was exposed to 50% glycerol for a period of 6 weeks. Although the pus no longer produces the disease (Hellerstrom and Wassén³), the glycerol does not reduce its antigenic activity. This provides another means of preservation of the antigen. The use as antigens of whole pus or the separate lipid and protein fractions failed to show complement fixation when set up with sera from patients with Lymphogranuloma inguinale.

After 48 hours sections of the skin of a positive Frei (1:100) reactor injected at the same time with (1:5) chancroidal pus showed infiltrations of lymphocytes around vascular buds, but no eosinophiles. The reaction was more marked in the case of the Frei antigen.

¹ Reiss, F., *Arch. Dermat. and Syph.*, 1935, **31**, 215.

² Levaditi, H., and Reinié, J., *Comptes rendus Soc. Biol.*, 1935, **118**, 123.

³ Hellerstrom, S., and Wassén, E., *Epidemiology and Etiology of Lymphogranuloma inguinale*, Special volume, dedicated to Prof. Cantacuzène-Masson Cie, 1934.