these fragments was similar to the 3-6s tryptic fragment. The other fragment (pap-Fc) had only specific IgM determinants that were partially deficient. None of these determinants were common to those on the 6-4s tryptic fragment. A peptide 'map' showed that only μ -chain peptides were present.

Treatment of IgM with pepsin gave very similar fragments to the tryptic ones. Small amounts of a fragment (pep-Fc) having only specific IgM determinants were also obtained. This fragment had more determinants than the pap-Fc fragment and some of these were common to those on the 6-4s tryptic fragment.

These results are consistent with a sub-unit structure for IgM; each sub-unit having a Fab portion containing a light chain and part of the heavy chain, and a Fc portion containing the rest of the heavy chain.

- Lamm, M. E. & Small, P. A. (1966). *Biochemistry*, **5**, 267.
 Miller, F. & Metzger, H. J. (1965a). *J. biol. Chem.* **240**, 3325.
 Miller, F. & Metzger, H. J. (1965a). *J. biol. Chem.* **240**, 4740.
 Miller, F. & Metzger, H. J. (1966). *J. biol. Chem.* **241**, 1732.
 Onoue, K., Kishimoto, T. & Yamamura, Y. (1967). *J. Immunol.* **98**, 303.

The Labelling of Brain Phosphatidylethanolamine and Ethanolamine Plasmalogen from Cytidine Diphosphate Ethanolamine *in vitro*

By G. B. ANSELL and R. F. METCALFE (*Department of Experimental Neuropharmacology, The Medical School, University of Birmingham*)

The formation of choline plasmalogen from 2-acyl 1-alk-1'-enyl glycerol (plasmalogenic diglyceride) and CDP-choline has been demonstrated by Kiyasu & Kennedy (1960) in the presence of a liver particulate fraction and very recently by Poulos, Hughes & Cumings (1968) with a particulate fraction from cardiac muscle. Both McMurray (1964a) and Ansell & Metcalfe (1966) have shown that labelling of both phosphatidylethanolamine and ethanolamine plasmalogen of brain from CDP-ethanolamine occurs when homogenates are incubated in the absence of an exogenous lipid acceptor.

When brain tissue (10mg.) from 20-day-old rats was incubated in 60mm-tris-HCl buffer, pH 8.0, for 1 hr. at 39° with 0.8mm-CMP-[³²P]phosphorylethanolamine (CMP-³²PE), the incorporation of [³²P]phosphorylethanolamine into the ethanolamine plasmalogen was stimulated sevenfold by the addition of 8mm-acyl alkenyl glycerol. This was a much greater stimulation than that found by McMurray (1964b). The activity of this enzyme, ethanolamine phosphotransferase (EC 2.7.8.1) to-

wards acyl alkenyl glycerol and diacyl glycerol in brain subcellular fractions was then determined. On the basis of both fresh brain weight and protein content, the activity was highest in the microsomal fraction, but significant activity was also found in a fraction consisting mainly of small myelin and synaptosomes, and also in the supernatant; less than 5% of the total activity was present in mitochondria. The distribution of activity towards diacyl and acyl alkenyl glycerol was very similar in all fractions examined. Subsequent experiments with the microsomal fraction showed that the K_m for acyl alkenyl glycerol was 1.7mm and that for diacyl glycerol 1.5mm, in the presence of saturating concentrations of CMP-³²PE.

For determining the activity of the enzyme in brain microsomes from animals of different ages this fraction (0.5-2mg. of protein) was incubated at 39° for 30min. in a volume of 1ml. containing: 60-80mm-tris-HCl, pH 8.0, 1.0-1.2mm-CMP-³²PE, 10mm-MnCl₂, and 0.03% Tween 20. In the absence of exogenous lipid substrate, the formation of ethanolamine plasmalogen was less than 1 μ mole/mg. of protein/hr. and that of phosphatidylethanolamine approx. 3 μ mole/mg. of protein/hr. At a saturating concentration of acyl alkenyl glycerol (10mm), the formation of ethanolamine plasmalogen was 70 μ mole/mg. of protein/hr., and at a saturating concentration of diacyl glycerol (15mm), the formation of phosphatidylethanolamine was 260 μ mole/mg. of protein/hr. Maximal activity towards substrates was between 16-30 days *post partum*. The variation in transferase activity towards acyl alkenyl glycerol during development was very similar to that observed towards diacyl glycerol. At 3 days *post partum*, ethanolamine phosphotransferase activities towards diacyl and acyl alkenyl glycerol were 58 and 56% respectively of the peak values at 16 days. The corresponding figures for the adult animals were 69 and 62% respectively of the peak values.

- Ansell, G. B. & Metcalfe, R. F. (1966). *Biochem. J.* **98**, 22P.
 Kiyasu, J. Y. & Kennedy, E. P. (1960). *J. biol. Chem.* **235**, 2590.
 McMurray, W. C. (1964a). *J. Neurochem.* **11**, 287.
 McMurray, W. C. (1964b). *J. Neurochem.* **11**, 315.
 Poulos, A., Hughes, B. P. & Cumings, J. N. (1968). *Biochim. biophys. Acta*, **152**, 629.