Rat brains allowed to autolyze in situ for 12 h selectively lost myelin proteins. Basic proteins are markedly decreased, but DM-20 (1) and proteolipid proteins also are lost from the myelin of developing and mature rat brain. At room temperature there is a 50% decrease in the concentration of basic protein in myelin isolated from 15-day-old rats. Reducing the ambient temperature to 0°C reduces the loss to 20%. Similar but less marked changes occur in the brains of adult animals. The molar ratios of cholesterol, phospholipids, and glycolipids are unaffected by autolysis, and there is also no change in the specific activity of the myelin marker enzyme 2',3'-cyclic nucleotide-3'-phosphohydrolase (E.C. 3.1.4.37). Electron microscopic examination of the isolated myelin demonstrates multilamellar structure with intraperiod lines.

INTRODUCTION

The effects of autolytic breakdown on biochemical constituents characteristic of central nervous system myelin have been investigated. Sammeck and Brady (2) observed that rat brain myelin basic proteins underwent rapid proteolysis within a few hours after death when the tissue was incubated in vitro at room temperature. Swanson et al. (3) studied subcellular fractionation of post mortem guinea pig brains left in situ at room temperature and found little change in the enzymatic activity and total protein content of the myelin fraction, isolated...
according to a modification of the technique of Gray and Whittaker (4). However, in this study the changes in individual proteins of central nervous system myelin were not examined. Ansari et al. (5) studied the effect of autolysis on the myelin basic protein of bovine cerebral hemispheres. They found that incubation of brain tissue at 23°C for varying periods of time produced no qualitative changes in the electrophoretic pattern of myelin basic protein in a phenol–formic acid–water gel system or on the quantity of basic protein isolated from the cerebral hemispheres unless the neural tissue had been frozen and thawed. The latter condition markedly reduced the concentration of myelin basic protein. Matthieu et al. (6) also did not find any change in the concentration of basic protein in adult bovine or mouse brains kept at 19°C postmortem but did find a significant decrease in basic protein concentration in myelin isolated from 25-day-old mouse brains under the same conditions. These results are in contrast to the observations of Sammeck and Brady (2). The present study was designed to investigate the effects of postmortem autolysis in situ on typical biochemical constituents of central nervous system myelin in developing and adult rat brain. In this communication we will present evidence that selective losses of major myelin proteins, especially basic proteins, and also proteolipid protein and DM-20 (1) occur in both age groups. There is no effect on the relative concentrations of the major lipid classes or the enzyme, 2',3'-cyclic nucleotide-3'-phosphohydrolase of myelin. A preliminary report of this work has been presented by Fishman et al. (7).

**EXPERIMENTAL PROCEDURE**

*Tissues.* Developing (15-day-old) and adult (65 to 70-day-old) Sprague–Dawley rats were sacrificed by the intraperitoneal injection of sodium phenobarbital. The animals were divided into three groups. In the first, myelin was isolated immediately upon death and hereafter is referred to as fresh. In the second group, the animals were kept at 0°C for 12 h, the brains were then removed from the cranial cavity, and myelin was isolated. The third group was kept at room temperature (25°C) for 12 h prior to removal of the brain and isolation of myelin.

*Chemicals.* All the agents used were analytical grade unless otherwise specified. The source of chemicals for sodium dodecyl sulfate gel electrophoresis and enzyme assays has been described previously by Agrawal et al. (8).

*Procedures.* Myelin was isolated as described previously (1). Lipids, total proteins, and the activity of 2',3'-cyclic nucleotide-3'-phosphohydrolase were measured as described previously by Fishman et al. (9).

*Gel Electrophoresis.* Myelin proteins from which lipids had been removed were solubilized in 5 mM sodium phosphate buffer (pH 7.2) containing 1% (w/v) sodium dodecyl sulfate, 8% (w/v) sucrose, and 1.5% (w/v) dithiothreitol. Proteins solubilized in sodium