LETTERS TO THE EDITOR

On the Chemical Structure of Lysolecithins

SIR,—For some years the action of phospholipids, especially of lysolecithins, on the cardiac muscle has been the subject of pharmacological studies in our institute. Therefore we have found the excellent Review Article on lysolecithin by N. Robinson (1961) in this Journal of great use to us. It seems to be of general value to report on recent results concerning the chemical structure and composition of lysolecithins.

When lecithin is attacked by lecithinase A, one fatty acid ester linkage is cleaved with the formation of lysolecithin and a long-chain fatty acid. In the past several attempts have been made to determine whether the α'- or the β-ester linkage in lecithin is attacked by lecithinase A. The problem seemed finally having been solved by Hanahan (1954) and Long and Penny (1954), who showed fairly conclusively that lecithinase A attacks only the α'-ester linkage. Thus it has been assumed that the fatty acid ester linkage in lysolecithin is in the β-position of the glycerol nucleus only (β-acyl lysolecithin).

More recent investigations by Marinetti and his colleagues (1959a) resulted in the presumption that lecithinase A is not specific for cleaving the α'-ester linkage only, since their lysolecithin, obtained by incubation with cobra venom, had also an α'-acyl structure. Later Tattrie (1959) was able to show that lecithin from egg yolk had only saturated fatty acids on the α'-position and unsaturated ones on the β-ester linkage of lecithin. The lysolecithin resulting from the incubation of such a lecithin with phospholipase A (from the venom of *Crotalus adamanteus*) contained only saturated fatty acids, as has been shown by gas chromatographic analysis. From this Tattrie concluded that lecithinase A cleaves the unsaturated β-linked fatty acids specifically. With this opinion, De Haas and van Deenen (1960) have agreed. They demonstrated by means of synthesised "mixed-acid" lecithins that lecithinase A splits exclusively the fatty acids attached to the β-position irrespective of whether they were saturated or unsaturated. Meanwhile, Hanahan and his colleagues (1960) have corrected their former results. In the summary of their recent publication they state: "Thus it seems that lecithinase A attacks specifically the β-ester position of lecithins forming α'-acyl lysolecithin."

Another way to form lysolecithin is the acid hydrolysis of plasmalogens. Prepared thus, lysolecithin is proabably not exclusively α'-acyl lysolecithin as described by Gray (1958), for Marinetti and his colleagues (1959b) reported, that in at least 68 per cent of all plasmalogens of beef heart muscle the fatty aldehyde residue was found to be attached to the α'-position. Therefore the acid hydrolysis of natural plasmalogens yields to a certain degree (up to 68 per cent for the beef heart muscle) β-acyl lysolecithin with a corresponding share of unsaturated fatty acids.

Institut für Pharmakologie,
Toxikologie und Pharmazie
der Tierärztlichen Fakultät
der Universität München,
Munich,
Germany.

A. PETTER.
W. SCHLEMMER.
K. ZIPF.

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Peroxide-Sulphuric Acid Test as an Indication of the Ripeness and Physiological Activity of Cannabis Resin

SIR,—Duquénois and Negm (1938) have reported a reaction with hydrogen peroxide and sulphuric acid as a very sensitive but not specific enough test for the identification of cannabis. This should not be mistaken for the "Duquénois-Negm" vanillin-acetaldehyde test for cannabis, described in the same paper, which has found a wide application for the identification of hemp resin. No further data on the use of the peroxide-sulphuric acid test for cannabis have been made available since.

To examine the reaction of various types of cannabis resin, 49 samples originating from 11 countries have been analysed by means of the following procedure. Cannabis was extracted by maceration in light petroleum (1 : 20)

TABLE I

Origin	Number of samples analysed	Ripeness	Colour obtained
Germany	5	Unripe	Pink
Switzerland	1	"	"
Yugoslavia	2	Intermediate	"
Morocco	1	"	"
Spain	4	"	Pink to reddish-brown
Cyprus	4	"	"
Greece	6	"	Reddish-brown to brown
Brazil	15	Ripe	Mostly brown
Costa Rica	1	"	Brown
Burma	1	Overripe	Greenish-brown
Cyprus	1	Spoiled	None
Canada	8	"	"

for 24 hr. 0·2 ml. of the extract was left to evaporate in a porcelain dish. To the residue 2 drops of a 20 per cent hydrogen peroxide and 0·5 ml. of concentrated sulphuric acid were added and the dish rotated gently for 1 min. The colour of the liquid was observed after 5 min.

Most samples gave colours which ranged from pink (temperate regions) to brown or greenish brown (mostly tropical samples). But some of the samples did not exhibit any colour at all. The results are summarised in Table I. For comparison, their ripeness has been recorded in the same Table. Full details of constants, based upon an indophenol reaction, are given elsewhere (Grlić, 1961).

To elucidate the differences obtained, the same test was made with pure cannabinolic compounds. Cannabidiol yields a pink product, which in higher concentration appears blood-red. Tetrahydrocannabidiol exhibits a violet colour. Six synthetic tetrahydrocannabinol homologues showed strong brown colours. Synthetic cannabiol showed at first a green colour going quickly