In the same manner simultaneous incubates containing only enzyme extract and ^{14}C methionine were assayed for methylhistamine. Now the carrier contained no radioactivity after only two crystallizations. It was thus concluded that carrier methylhistamine can, by repeated crystallization of the dipicrate, be completely freed from contamination by ^{14}C methionine and its possible metabolites.

The destruction of histamine and the formation of methylhistamine was followed by simultaneous incubation of the crude enzyme extract with radiocarbon histamine labelled in the imidazole nucleus. At the end of the incubation remaining histamine and methylhistamine were assayed by isotope dilution technique. It has been shown that by repeated crystallization of the dipicrate carrier amounts of methylhistamine can be freed from contamination by ^{14}C histamine or its metabolic products. By conversion of carrier histamine to its pipsyl derivative the contaminating methylhistamine can be removed [7]. Therefore constant activity of the samples during three successive crystallizations was considered as a sufficient criterion of the purity.

The essential figures from an experiment are given in Table 1. It has been concluded from these results that the methyl group of methylhistamine can be transferred from methionine. Whether methionine is the immediate precursor of the methyl group or an intermediate step is involved cannot be concluded from this type of experiment.

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Phosphorus-32 Pentasulfide: Preparation by Isotopic Exchange and Conversion to Thiophosphoryl-32 Chloride and Phosphorus-32 Trichloride

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Certain triesters of phosphoric and thiophosphoric acids are important as pesticides and potential chemical warfare agents. Adequate studies on the metabolism and mode of action of these organophosphorus compounds require their synthesis from high specific activity radiolabeled inorganic intermediates such as phosphorus pentasulfide (P_2S_5), thiophosphoryl chloride ($PSCl_3$), phosphorus trichloride (PCl_3), and phosphoryl chloride ($POCl_3$).

$^{32}POCl_3$ can be readily prepared in high specific activity from the reaction of $H_3{}^{32}PO_4$ with phosphorus pentachloride (PCl_5)[1]. Neutron irradiation is commonly employed to label certain of the other inorganic phosphorus intermediates [2-5]. This method is often inconvenient and the specific activity of the organophosphorus compounds prepared from these irradiated intermediates is usually not greater than 5 mC/g. $^{32}PSCl_3$ can also be prepared from $H_3{}^{32}PO_4$ *via* $^{32}POCl_3$ and $^{32}PCl_3$ [6,7] or by an inefficient isotope exchange reaction [8].

Various procedures were tried to achieve isotope exchange between $H_3{}^{32}PO_4$ or $Na_2H^{32}PO_4$ and red phosphorus, $PSCl_3$ or P_2S_5. The products from each attempt were reacted with ethanol [9] or converted to other known organic derivatives [4] and then separated by paper and ion-exchange chromatography [10] to determine the degree of exchange. After correction for the radioactivity remaining on the glassware, the

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only significant exchange occurred with the sulfur-containing compounds. The percentage exchange was low with $PSCl_3$ but almost complete with P_2S_5.

For preparation of $^{32}P_2S_5$, carrier-free $H_3{}^{32}PO_4$ or $Na_2H^{32}PO_4$ was evaporated in a reaction flask. The acid was preferable when the residue after evaporation was high and the salt when the residue was no more than a few milligrams. Red phosphorus and sulfur were then added in a 2 to 5 molar ratio with a combined weight of 0.20 to 2.0 g. While gassing the flask with dry carbon dioxide, the mixture was heated in one spot with a weak flame to initiate the vigorous reaction[11] that liquified the mixture and formed P_2S_5. When the mixture was then boiled for 10 min the exchange reaction was over 98 % complete. Similar exchange (95—98 %) resulted by slowly melting P_2S_5 in a reaction flask and then boiling for 10 min while gassing with carbon dioxide, and by heating the P_2S_5 for 2 h at 300—500°C in sealed ampules. The specific activity of the $^{32}P_2S_5$ thusly formed remained constant through repeated recrystallizations from carbon disulfide. When this $H_3{}^{32}PO_4$—P_2S_5 exchange reaction was run in the presence of varying amounts of non-labeled H_3PO_4, the percentage of the radioactivity recovered as $H_3{}^{32}PO_4$ closely approximated the percentage of the total phosphorus in the reaction mixture contributed by the H_3PO_4.

$^{32}PSCl_3$ was prepared in 73 % yield from $^{32}P_2S_5$ by reaction with 3 molar equivalents of PCl_5[12,13] in the presence of anhydrous aluminium chloride comparable to 1.5 times the weight of the sulfur. The reaction was carried out in a closed all-glass system for 1.5 h at 160°C. $^{32}PCl_3$ was then formed in 91 % yield by reaction of the $^{32}PSCl_3$ with 1.1 molar equivalents of triphenyl phosphine at 130—140°C for 2 h according to a known type reaction[14] for obtaining sulfur migration from $PSCl_3$.

$^{32}P_2S_5$ prepared by this isotope exchange reaction has been used in the preparation of 15 different organic phosphate and phosphorothioate insecticides. Yields of the chromatographically-pure insecticides varied from 20—90 % based both on total phosphorus and radioactivity. The specific activities ranged from 2 to 340 mC/g. A more detailed report on the conversion of the $^{32}P_2S_5$ to various labeled insecticides and the characterization of the radioactive products will be published elsewhere.

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