2. Evidence is presented for a regular variation in the specific adenosine triphosphatase activity of myosin fractions eluted from diethylaminoethyl-cellulose.

3. Partial separation of 5'-adenylic deaminase activity has been obtained by chromatography at pH 8.2.

4. Myosin has been prepared with less than 0.002% of phosphorus and virtually free from the ribonucleic acid invariably present in L-myosin preparations.

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Distribution and Activity of Monoamine Oxidase in Mouse Tissues

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During a recent study of the radioprotective action of 5-hydroxytryptamine in the mouse (Hope, 1959) we were impressed by its low toxicity in this species compared with the rat. The LD₅₀ for 5-hydroxytryptamine administered intraperitoneally as the creatinine sulphate in the mouse was approximately 1 g./kg. body weight. Rapport & Virno (1952) found that the LD₅₀ for this substance administered in the same way in rats was 50 mg./kg. body weight, i.e., it is 20 times as toxic to the rat as to the mouse.

Rapport & Virno (1952) investigated in a number of mammalian species the fall in oxygen consumption after the administration of 5-hydroxytryptamine. The rat was more sensitive in this respect than any other species examined. The authors

suggested that the low amine oxidase activity of rat kidney was responsible.

In view of the now well-established fact that amine oxidase is an important catalyst of the destruction of 5-hydroxytryptamine in mammals (Blaschko, 1958) we decided to study the activity of the enzyme in mouse kidney and some other mouse tissues.

In this paper the rate of oxidation of 5-hydroxytryptamine has been compared with that of representatives from the main types of monoamine oxidase substrates. These included tryptamine, β -phenylethylamine, isoamylamine and benzylamine. Tyramine, although in some respects an unsatisfactory substrate of amine oxidase (Blaschko & Himms, 1954), was also included in order to

compare the results obtained with those of other workers.

The amine oxidase of mouse kidney has been shown to have a high degree of specificity for 5-hydroxytryptamine; this amine was the most rapidly oxidized substrate. This finding was followed by a study of four other tissues: brain, liver, lung and spleen. The presence of an amine oxidase in mouse liver has previously been described by Kobayashi (1957). This enzyme possesses many of the properties of monoamine oxidase yet it also attacks histamine and cadaverine. Davison, Lessin & Parkes (1957) have demonstrated the presence of tyramine oxidase in mouse liver and brain. Brain amine oxidase is worthy of special note because of the current interest in the functions of amines in this organ.

The effects of the amine oxidase inhibitors amphetamine [(+) and (-)] and iproniazid on the enzymic activity of mouse tissues have been investigated. Use has been made of homogenates of brain tissue from mice treated with iproniazid in a study of the spontaneous oxidation of 5-hydroxytryptamine during the determination of the optimum substrate concentration for this compound.

Since guanidines are known to be inhibitors of monoamine oxidase an attempt to produce an inhibitor combining the high specificity of the 5-hydroxyindole residue with a guanidine was made. 5-Hydroxytryptacyamine, the guanidine analogue of 5-hydroxytryptamine, was synthesized and isolated as its creatinine sulphate. The inhibitory action of this substance on amine oxidase was measured.

MATERIALS AND METHODS

Substances. The amines isoamylamine, benzylamine and β -phenylethylamine as their hydrochlorides and 5-hydroxytryptamine creatinine sulphate monohydrate were obtained from Roche Products Ltd. Tryptamine and tyramine hydrochlorides were obtained from British Drug Houses Ltd.

The optical isomers of amphetamine hydrochloride were kindly given by Dr H. Blaschko. The resolution was effected by Professor P. Pratesi. We are also grateful to Dr A. L. Morrison (Roche Products Ltd.) for a gift of iproniazid phosphate.

Animals. The mice used were females of the R strain used in this Department [a cross between PCT ϕ and (C3H $\times$ 101) σ]. The animals were 14 weeks old when killed; their mean weight was 24.5 g.

Treatment with iproniazid. Iproniazid (*N*-isonicotinoyl-*N'*-isopropylhydrazine) as the phosphate salt was administered intraperitoneally in a neutral 0.9% NaCl solution.

Preparation of tissue extracts. The animals were killed by a blow on the head followed by decapitation. Tissues from 30 or 40 animals were pooled and homogenized in an all-

glass Potter-Elvehjem homogenizer with 4 vol. of 0.067M-sodium phosphate buffer, pH 7.4. The homogenates were dialysed against distilled water (500 ml.) with three changes over a period of 18 hr. at 2° in order to reduce the blank consumption of O₂. This was usually 0.02–0.05 μ l./mg. of fresh tissue/hr.

Measurement of enzymic activity. The enzymic activity was determined manometrically. The main compartment of the manometer flask contained 1 ml. of the dialysed homogenate and 0.6 ml. of 0.2M-sodium phosphate buffer, pH 7.4. Where inhibitors were used, the buffer volume was reduced to 0.4 ml. and the inhibitor in a volume of 0.2 ml. was present in the main compartment. The system used for determining the rate of consumption of O₂ in the presence of cyanide was that devised by Krebs (1935). The side bulb contained 0.4 ml. of water or of 0.05M-solution of the amine to be tested; the inner well contained 2N-KOH and filter paper. The gas phase was O₂ and the temperature 37.5°.

The enzymic activity was expressed as q_{O_2} , i.e. as μ l. of O₂ taken up/mg. of fresh tissue/hr.; it was calculated from the initial period when the rate of reaction was approximately constant.

5-Hydroxytryptamine (5-HT) was included as a substrate in each experiment and its rate of oxidation was compared with that of the other amines selected.

Preparation of the guanidine analogue of 5-hydroxytryptamine. 5-Hydroxytryptacyamine was synthesized from 5-HT creatinine sulphate by reaction with *S*-methyl-isothiuronium sulphate. The reaction of amines with thiuronium salts for the synthesis of guanidines is well known.

5-HT creatinine sulphate monohydrate (1 g.) and *S*-methyl-isothiuronium sulphate (2.19 g.) were dissolved in water (6 ml.). Ammonia was added until the mixture was strongly alkaline (3 ml. of aq. NH₃ soln., sp.gr. 0.880), and the temperature was maintained at 60–70° for 2 hr. The reaction occurred with evolution of methyl mercaptan. A further 2 ml. of aq. NH₃ soln. was added and the mixture boiled for 10 min. After acidification with 5N-H₂SO₄ an equal volume of ethanol was added. A crystalline product rapidly separated in the cold. This product (0.65 g.) had m.p. 285–286°. Recrystallization was effected from water. The product separated in fine needles and had m.p. 287–289°. Elementary analysis showed that the product was 5-hydroxytryptacyamine creatinine sulphate (Found: C, 41.7; H, 5.57; N, 22.8; S, 7.35; C₁₅H₂₃O₄N₃S requires C, 41.95; H, 5.40; N, 22.83; S, 7.47%). Paper chromatography in butan-1-ol-acetic acid-water (4:1:5; upper phase) on Whatman no. 4 paper gave a single spot having R_F 0.48. The position of the substance was revealed as a pink spot on the chromatograms by spraying with a mixture made as follows: diaecetyl (1%, v/v, in water; 2.5 ml.) with α -naphthol (25%, w/v, in propan-1-ol; 20 ml.) was made up to 100 ml. with propan-1-ol. Just before use, 10 ml. of this solution, together with 5 ml. of 3N-NaOH, was made up to 30 ml. with water to give the spray reagent. This system was developed from the method devised by Rosenberg, Ennor & Morrison (1956) for the estimation of guanidines.

RESULTS

The results of the substrate-specificity studies are summarized in Table 1. The result of one experiment with each substrate and tissue has been included. With brain and kidney the experiments

Table 1. *Relative rates of oxidation of 5-hydroxytryptamine and other amines by homogenates of mouse tissues*

Initial substrate concentration: 10 mM. Gas phase, O₂; temp., 37.5°. The q_{O_2} (μ l. of O₂ consumed/mg. of fresh tissue/hr.) with 5-HT as substrate is given in parentheses.

Tissue	Substrate					
	5-Hydroxy-tryptamine	Tryptamine	Tyramine	β -Phenylethylamine	<i>iso</i> Amylamine	Benzylamine
Brain	100 (0.490)	49	155	41	38	25
Liver	100 (0.695)	147	388	135	126	105
Kidney	100 (0.575)	47	77	26	0	0
Spleen	100 (0.248)	60.5	85	60.5	0	0
Lung	100 (0.750)	35	—	59	52	42

Fig. 1. Oxygen uptake due to oxidation of amines by mouse-kidney homogenate. Each flask contained: kidney homogenate equivalent to 110 mg. of fresh tissue (1 ml.); 0.2M-sodium phosphate buffer, pH 7.4 (0.6 ml.). The substrates were present at initial concentrations of 0.01M. Final volume, 2 ml. KOH was present in the centre well. Gas phase, O₂; temp., 37.5°. ○, 5-Hydroxytryptamine; ■, β -phenylethylamine; ●, tryptamine; □, tyramine.

have been repeated several times and the results confirmed. The activity of amine oxidase is constant in these two tissues. In four experiments with brain homogenates and 5-HT as substrate the q_{O_2} ranged from 0.46 to 0.49; for kidney the corresponding figures were 0.55 to 0.57.

Kidney. 5-HT was the amine most rapidly oxidized by homogenates of mouse kidney. Tryptamine, tyramine and β -phenylethylamine were also destroyed but at slower rates. *iso*-Amylamine and benzylamine do not appear to be substrates for the kidney enzyme; this distinguishes the kidney enzyme from those present in brain and liver. The kidney enzyme closely resembles the brain enzyme in the relative rates of oxidation of the other substrates, e.g. β -phenylethylamine was oxidized more slowly than tryptamine. The introduction of a hydroxyl group into the aromatic ring systems of these substances greatly increases the oxidation rates. Thus it can be seen in Fig. 1 that tyramine and 5-HT are the optimum substrates.

Brain. Homogenates of mouse brain oxidize 5-HT at about twice the rate for tryptamine or for β -phenylethylamine. The most rapidly oxidized substrate was tyramine. *iso*Amylamine and benzylamine were also oxidized, at slower rates.

Concentrations of 5-HT from 0.625 mM to 0.16M were used to study the effect of substrate concentration on the reaction rate. The result of this experiment is shown in Fig. 2. The rate of oxidation increases linearly with the logarithm of the concentration of 5-HT up to 7.5 mM. At this point there is a discontinuity; the rate continues to increase but more slowly. The increase in rate beyond the discontinuity is due almost entirely to a spontaneous oxidation of 5-HT at high concentrations in an atmosphere of O₂. The lower curve in Fig. 2 shows the relationship between the concentration of 5-HT and the oxidation rate when homogenate prepared from the brains of mice treated with iproniazid was used. Two doses of iproniazid (100 mg./kg. body wt.) were given at 24 hr. intervals in order to inhibit fully the amine oxidase activity (see below). The homogenate from mice treated with iproniazid was used as a control because the rate of spontaneous oxidation of 5-HT is slightly higher in the absence of tissue proteins. The spontaneous rate cannot be measured manometrically when 5-HT concentrations below 0.01M are used.

It seems reasonable to assume from this experiment that the optimum substrate concentration is approximately 0.01M. The concentration for 50% of the maximum rate is 1.25 mM. This result may be compared with that obtained by Kohn (1937), who found the Michaelis constant for tyramine and the pig-liver amine oxidase to be 0.5 mM.

Fig. 2. Effect of 5-HT concentration on the oxidation rate with brain homogenates from normal (O) and iproniazid-treated (●) mice. Manometer flask contents: dialysed homogenate (1 ml.) derived from 300 mg. of fresh brain tissue; 0.2M-sodium phosphate buffer, pH 7.4 (0.6 ml.); suitable substrate concentrations were present in the side arm to give the initial concentrations shown in the figure. Gas phase, O_2 ; temp., 37.5° .

Fig. 3. Effect of pH on the rate of oxidation of 5-HT by mouse-brain homogenate. Each flask contained brain homogenate equivalent to 170 mg. of fresh tissue (1 ml.) and 0.2M-sodium phosphate buffer of requisite pH (0.6 ml.). 5-HT was present in an initial concentration of 0.01M. Final volume, 2 ml. Gas phase, O_2 ; temp., 37.5° .

In a similar experiment with tryptamine the rate of consumption of O_2 was constant when the substrate concentrations varied between 0.625 and 20 mM. This at first suggested that the optimum substrate concentration for tryptamine was lower than for 5-HT. However, when the reaction was taken to completion the consumption of O_2 was four times larger than could be expected from the following reaction scheme:

Much of the extra oxygen consumption could be prevented by the addition of cyanide (10 mM) in the manometric experiments. This showed that enzymes other than the cyanide-insensitive amine oxidase were involved. The percentage inhibition of uptake of O_2 by cyanide was 10, 30 and 50 at tryptamine concentrations of 10, 5 and 2.5 mM respectively. The full significance of this result is not understood; it may be that the secondary oxidation of the product of amine oxidase action is sensitive to concentration. This complication rendered it impossible to estimate the Michaelis constant without purifying the enzyme.

The optimum pH for the brain enzyme was 8.1. The activity-pH curve is shown in Fig. 3.

Liver. In mouse-liver homogenates tyramine was the most rapidly oxidized substrate. The enzyme destroyed each of the substrates tested at appreciable rates; the lowest rate was observed with 5-HT. Thus the liver stands in marked contrast with the other tissues studied.

Other tissues. Lung and spleen tissue were also examined for enzymic activity. The highest q_{O_2} value for 5-HT oxidation was found with lung homogenates. 5-HT was moreover the most readily oxidized substrate.

Spleen tissue showed a relatively weak activity; 5-HT was again the most readily attacked substrate. Homogenates of spleen resembled those from kidney because neither *isoamylamine* nor *benzylamine* was oxidized.

A comparison of (+)- and (-)-amphetamine as inhibitor of amine oxidase in brain and kidney. 5-HT has been used as the substrate in a comparison of the optical isomers of amphetamine as inhibitors of monoamine oxidase activity in mouse-brain and kidney homogenates. A range of concentrations of the isomers was incubated with enzyme in the main compartment of the manometer flask during equilibration.

It can be seen in Fig. 4 that in the brain (+)-amphetamine was seven times as active as (-)-amphetamine. In kidney (+)-amphetamine was also the more active isomer but the difference observed was much less than that observed with the brain enzyme. (-)-Amphetamine is equally as

effective in the kidney as in the brain. The concentrations (mM) of the isomers necessary to inhibit the enzymic activity by 50 % were:

	(+)-Amphetamine	(-)-Amphetamine
Brain	0.513	3.63
Kidney	1.51	3.63

Inhibition of amine oxidase by administration of iproniazid. Davison *et al.* (1957) found that the amine oxidase activity in brain and liver was

completely inhibited in mice 24 hr. after the intra-peritoneal administration of iproniazid (100 mg./kg. body wt.). The amine oxidase activity of the R strain of mice was more resistant to iproniazid than that of the albino mice used by Davison *et al.* After a second administration the brain enzyme became fully inhibited, yet the enzyme in the liver seemed not to be affected. From 10 to 20 % of normal activity was retained with all substrates in liver. This was also true in kidney, except for β -phenylethylamine, where oxidation appeared to be fully inhibited by two injections of iproniazid. This may be due to an experimental error arising from the difficulty of measuring accurately rates of oxidation of the same order of magnitude as the blank O_2 consumption. The results of these experiments can be seen in Table 2.

Experiments with 5-hydroxytryptacyamine. 5-Hydroxytryptacyamine was not oxidized when incubated with homogenates of mouse brain in concentrations up to 10 mM.

The inhibitory activity against the brain enzyme with 5-HT as substrate was relatively weak. The concentration of the guanidine necessary to inhibit the enzymic activity by 50 % was 8.9 mM.

DISCUSSION

Differences in substrate specificity as well as in the relative rates of oxidation of the amines suggest the possibility that each tissue may contain a distinct enzyme for the destruction of monoamines.

In all the tissues examined tryptamine was oxidized more slowly than tyramine. Schayer, Wu, Smiley & Kobayashi (1954) concluded that in the mouse tryptamine was metabolized at a rate approximately 30 % above that of tyramine. This apparent contradiction may be explained if tryptamine has a higher affinity for amine oxidase than tyramine. This is not yet known for the mouse though this explanation holds for the rat, where a similar situation exists. Rat-liver amine oxidase has a greater affinity for tryptamine than

Fig. 4. Inhibition of the amine oxidase from mouse brain and kidney by (+)- and (-)-amphetamine. x, Kidney with (-)-amphetamine; ●, kidney with (+)-amphetamine; □, brain with (-)-amphetamine; ○, brain with (+)-amphetamine. Manometer flask contents: dialysed homogenate (1 ml.) derived from 200 mg. of fresh tissue; the substrate, 5-HT, was present in each flask at an initial concentration of 10 mM; final volume, 2 ml. Inhibitor (0.2 ml.) was added to 0.2M-sodium phosphate buffer, pH 7.4 (0.4 ml.). Gas phase, O_2 ; temp., 37.5°.

Table 2. *Amine oxidase activity in tissues from mice treated with iproniazid*

Enzymic activity is expressed as a percentage of the normal. Injection 1 consisted of 100 mg. of iproniazid/kg. body wt. 24 hr. before killing the animals. Injection 2 consisted of two administrations of 100 mg. of iproniazid/kg. body wt. 48 and 24 hr. before killing the animals. Initial substrate concentration, 10 mM. Gas phase, O_2 ; temp., 37.5°. Actual q_{O_2} values (μ l. of O_2 consumed/mg. of fresh tissue/hr.) are given in parentheses.

Injection	Tissue	Substrate				
		5-HT	Tryptamine	β -Phenylethylamine	Tyramine	isoAmylamine
1	Brain	29 (0.14)	58 (0.14)	0 (0)	19 (0.14)	0 (0)
	Kidney	23 (0.13)	11 (0.03)	25 (0.04)	20 (0.09)	—
	Liver	15 (0.10)	10 (0.10)	12 (0.11)	21 (0.57)	19 (0.17)
2	Brain	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
	Kidney	20 (0.11)	26 (0.07)	0 (0)	18 (0.08)	—
	Liver	16 (0.11)	10 (0.10)	14 (0.13)	20 (0.54)	21 (0.18)

for tryamine, yet when these amines are tested singly tyramine is oxidized more readily than tryptamine. When the substrates are present together, tryptamine is metabolized faster (Kobayashi & Schayer, 1955). Langemann (1955) has shown that rat-liver mitochondria destroy 5-HT more rapidly than tryptamine yet only half as rapidly as tyramine. In heart-muscle mitochondria and myofibrils, on the other hand, 5-HT and tyramine are destroyed at similar rates. Tryptamine was in all cases less rapidly oxidized than 5-HT. Thus we see evidence in rats as well as mice for the view that in a number of tissues, if not all, amine oxidase has a high affinity for the indole amines and, more especially, for 5-HT.

5-HT is optically inactive; it was therefore surprising to find that the inhibition of amine oxidase activity in brain and kidney by amphetamine was greater with the (+)- than with the (-)-isomer. The brain enzyme was more optically specific than the kidney enzyme. P. Pratesi & H. Blaschko (1957, personal communication) showed that guinea-pig liver amine oxidase is more sensitive to inhibition by (+)-amphetamine than by (-)-amphetamine.

Pletscher & Gey (1958) have recently described two stereospecific amine oxidase inhibitors derived from the α -amino acids alanine and serine. The structures of these substances are:

In both the more active isomer *in vitro* and *in vivo* was that derived from the naturally occurring amino acid. Pletscher & Gey suggested that these differences could be partly ascribed to an optically active centre in the enzyme.

The finding that (+)-amphetamine is a considerably more potent inhibitor of mouse-brain amine oxidase than the (-)-isomer is of interest because of what is known of their effects on the central nervous systems of man and animals. Schulte, Reif, Bacher, Lawrence & Tainter (1941) studied the central stimulant activity of (+)- and (-)-amphetamine in un-narcotized rats. The (+)-isomer was three times as active on a weight basis as the (-)-isomer. Alles (1939) had previously shown by subjective observations in man that (+)-amphetamine was four times as active as (-)-amphetamine.

In their peripheral effects the isomers of amphetamine are quantitatively more alike, e.g. (+)- and (-)-amphetamine are approximately equally active in decreasing the propulsive motility of the small intestine in rats and dogs (Van Liere, Stickney, Northup & Bell, 1951). Possibly these findings are related to the observation made in the present work that brain amine oxidase displays more

optical specificity than the enzyme found in a peripheral tissue (kidney).

The results obtained with mice treated with iproniazid show some differences from those obtained by Davison *et al.* (1957). Neither the brain nor the liver enzyme was fully inactivated by a single dose of iproniazid (100 mg./kg. body wt.). Approximately 20% of the activity remained in each tissue and also in kidney. A second similar dose of iproniazid did not alter the activity in liver and kidney, but the brain enzyme was fully inactivated. The rate of oxygen consumption with tryptamine as substrate was less inhibited than with other substrates in brain homogenates from mice after a single treatment with iproniazid (see Table 2). This phenomenon is probably related to that described earlier (for inhibition by cyanide) and is due to the further metabolism of the product of amine oxidase action on tryptamine.

The reason for differences between these results and Davison's is not at present known; it is possible that a strain difference is responsible. The mice used in the present studies are characterized by a high degree of hybrid vigour not possessed by albino mice used by Davison.

SUMMARY

1. 5-Hydroxytryptamine, tryptamine, tyramine, β -phenylethylamine, *iso*amylamine and benzylamine have been tested as substrates of amine oxidase in homogenates of the following mouse tissues: brain, kidney, liver, lung and spleen.
2. Mouse kidney contains a monoamine oxidase which destroys 5-hydroxytryptamine more rapidly than tyramine, tryptamine or β -phenylethylamine. This is also true of lung and spleen.
3. 5-Hydroxytryptamine was oxidized most rapidly by lung.
4. In liver homogenates 5-hydroxytryptamine was oxidized less rapidly than any other monoamine tested, yet the rate was only slightly less than that observed with lung tissue.
5. *iso*Amylamine and benzylamine, although oxidized by liver, brain and lung, were not attacked by kidney and spleen.
6. Differences in substrate specificity and relative rates of oxidation of the substrates serve to distinguish the amine oxidase from each tissue.
7. (+)-Amphetamine was seven times as potent as (-)-amphetamine as an inhibitor of brain amine oxidase; the difference was less with kidney.
8. The guanidine analogue of 5-hydroxytryptamine was synthesized. It is a weak inhibitor of monoamine oxidase. These findings are discussed in relation to the function of amine oxidase.

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Studies on the Synthesis of Serum Albumin by the Isolated Microsome Fraction from Rat Liver

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Zamecnik & Keller (1954) were the first to show that when radioactive amino acids are incubated with the microsomal fraction from rat liver, together with the soluble cytoplasm and a source of continuously regenerated adenosine triphosphate, an incorporation of amino acid into microsomal protein occurs. Since then the steps in this process of incorporation have been extensively studied; the precise nature of the radioactive microsomal proteins has remained undefined, although some progress in this direction was made by Simkin & Work (1957) by extracting the microsomes with solutions of varied ionic strength and pH. It is therefore not clear to what extent this incorporation of radioactive amino acids into substances precipitated by cold trichloroacetic acid but insoluble in hot acid represents the synthesis of a complete protein. It was in the hope that an answer to this question might be found that the present experiments were initiated.

Experiments on the perfused rat liver indicate that this organ is able to synthesize serum albumin (Miller, Bly, Watson & Bale, 1951). Moreover, it is possible to demonstrate the synthesis of serum albumin in incubated slices of either chick liver (Peters & Anfinsen, 1950) or rat liver (Campbell & Stone, 1957a). This demonstration depended on the use of specific antisera by which means the net synthesis of serum albumin during the incubation period could be shown. Further, if the slices were incubated in the presence of a radioactive amino acid, the serum albumin became radioactive and the degree of incorporation of radioactivity was approximately correlated with the amount of

protein synthesized (Campbell & Stone, 1957a). Peters (1957) later showed that chick-liver microsomes contained a small amount of serum albumin which could be solubilized. It was therefore decided in the present experiments to incubate rat-liver microsomes in the presence of radioactive amino acids under the conditions described by Zamecnik & Keller (1954) and to determine whether the amino acids were incorporated into the serum albumin which could be released from the microsomes. It was assumed that it would not be practicable to demonstrate the net synthesis of serum albumin in such preparations.

The major difficulty in conducting these experiments has been to demonstrate unequivocally that the isolated radioactive protein has all the characteristics of serum albumin. As will be shown, immunological, electrophoretic and solubility criteria have been applied in an attempt to demonstrate this. The use of a wide variety of criteria is particularly important for serum albumin since this protein retains its immunological characteristics even though it has been modified, e.g. by acetylation (Marrack & Orlans, 1954), or split into smaller fragments (Lapresle, 1955; Porter, 1957).

Since the amount of albumin which can be released from the liver microsomes is very small, it has been necessary to take advantage of every procedure which would ensure the isolation of albumin with the highest possible radioactivity. It has previously been shown (Hultin & Decken, 1957; Decken & Hultin, 1958; Campbell & Greengard, 1959) that the rat-liver microsome fraction derived from rats after partial hepatectomy is much more