Accordingly, the gastric emptying time of the solute is prolonged. The solution is further diluted during transit through the small intestine. This interrelationship between gastrointestinal water transport, gastric emptying time and rate of intestinal absorption, might be of general importance.

Ca-b.s. do not cause morphological or irreversible functional alterations in the intestine.

The implications of these findings will be discussed in relation to the results of other workers.

66a Gesetzmässigkeiten des Strontium - Stoffwechsels und inre Bedeutung für die Eliminierung von Strontium aus dem Skelet. A. SCHMID (Deutschland).

Es wird über Gesetzmässigkeiten des Strontium-Stoffwechsels im Skelett, den Umsatzmechanismus *in vivo* und die Fixierung von Strontium im Knochensystem berichtet.

66b Patterns of Strontium Metabolism and their Significance in the Elimination of Strontium from the Skeleton. A. SCHMID (Germany).

The patterns of strontium metabolism in the skeleton, the conversion mechanism *in vivo* and fixing of strontium in the bone system are reported.

67 On the Use of Expiratory $^{14}CO_2$ Patterns as a Pharmacological Tool for Studying the Biochemical Effects of Drugs. G. T. OKITA (U.S.A.).

Since all carbon containing biochemical intermediates are eventually metabolized to CO_2, alteration in their metabolism due to biochemical effects of a drug may be reflected by alteration in the rate at which labelled CO_2 appears in expired air. Therefore, an apparatus which will monitor continuous expiratory $^{14}CO_2$ patterns after the administration of ^{14}C-labelled intermediates is a useful pharmacological tool for studying the mode of action of those drugs having biochemical effects as a basis for their pharmacologic response. An apparatus built in our laboratory for this purpose has been reported elsewhere.[1] Essentially, the instrument consists of a 4 π gas phase Geiger counter, an infra-red gas analyzer for measuring $^{12}CO_2$, a ratio analyzer to compute specific activity ($^{14}CO_2/^{12}CO_2$) of $^{14}CO_2$, and a ventilation meter. All measurements are recorded continously on a 4-channel recorder after the injection of a ^{14}C-labelled intermediate. Depending upon the drug under investigation such labelled intermediates as acetate, pyruvate, lactate, formate, glucose, citrate, etc. have been employed. By the use of appropriate labelled intermediates and comparison of expiration $^{14}CO_2$ patterns between control and drug treated groups, it is possible to obtained information on the biochemical mode of action of drugs.

The effects of testosterone, oestrogen, insulin, orinase and diamox on expiratory $^{14}CO_2$ patterns after the administration of various ^{14}C-labelled intermediates will be presented. Some of the advantages of this method for studying biochemical effects of drugs are: (1) *in vivo* condition, all experiments are conducted on intact, unanaesthetized animals and subjects; (2) simplicity, no individual $^{14}CO_2$ samples to assay; also, 2–6 experiments may be run per day; and (3) utilization of human subjects, therefore, no need to extrapolate animal data.

1. (1960), *Int. J. Appld. Rad. Iso.*, **7**, 273.

68 Metabolic Studies of Carcinogenesis Using Expiratory $^{14}CO_2$ Patterns Following Administration of ^{14}C-Labelled Intermediates. E. A. EZZ and G. T. OKITA (U.S.A.).

Using an instrument developed in our laboratory[1] we were able to measure continuously expiratory $^{14}CO_2$ patterns in experimental animals after the administration of the following ^{14}C-labelled intermediates: acetate-1-^{14}C, sodium bicarbonate-^{14}C, glucose-1-^{14}C and glucose-6-^{14}C. The effect of carcinogenesis and various hormonal states such as ovariectomy, estrogen and testosterone therapy on the expiratory $^{14}CO_2$ specific activity patterns of ^{14}C-labelled intermediates were studied in virgins, exbreeders and mammary tumour C3H mice free of the mammary tumour "milk factor". The specific activity curves as well as ^{14}C levels in expiratory CO_2 showed significant differences in some of the experimental conditions. The most striking biochemical change noted during the carcinogenesis process was the reduction in the percentage recovery of glucose-1-^{14}C/glucose-6-^{14}C. Rank order arrangement for the various experimental groups of C3H mice were as follows: factor free—1·48, virgin—1·36, exbreeders—1·00, tumour (single)—0·84, and tumour (multiple)—0·75. The decrease in the ratio is a reflection of an increase in glycolytic metabolism. Ovariectomy and testosterone therapy to tumour animals tends to return the ratios to those for virgin controls. This supports the thesis that ovariectomy and testosterone therapy tend to correct the metabolic defect produced by tumour.

1. (1960), *Int. J. Appld. Rad. Iso.*, **7**, 273.

69 Studies on the Functions and Mode of Action of Thiamine. C. J. GUBLER (U.S.A.).

Although the symptoms of thiamine deficiency have been well documented, the metabolic disturbances which cause these symptoms are still not well understood. In order to gain a better understanding to these metabolic disturbances, and thus of the physiological functions of thiamine, rats were made

deficient by thiamine-deprivation and by administration of the thiamine antagonists oxythiamine (OTh) and pyrithiamine (PTh). At the onset of symptoms, rats were sacrificed and the oxidative decarboxylation of various α-ketoacids studied in the tissues by a spectrophotometric method with $Fe(CN)_6$ as the electron acceptor. In liver mitochondria thiamine deprivation, OTh-treatment, and PTh-treatment all caused a reduction in the rate of oxidative decarboxylation of pyruvate (to 24·3, 46·2 and 43·1 per cent of the normal rate, respectively). The rate with α-ketoglutarate was affected only by thiamine-deprivation (66·2 per cent of normal). With α-ketoisovalerate and α-keto-β-methylvalerate, none of the deficiencies had a significant effect. In kidney homogenates all three types of deficiency caused a marked reduction in pyruvate oxidation (41·8, 44·3 and 37·9 per cent of normal, respectively) but only thiamine-deprivation and PTh-treatment caused a decrease in α-ketoglutarate oxidation (45·5 and 65·2 per cent, respectively). OTh had no effect. In brain homogenates PTh-treatment caused a decrease in both pyruvate and α-ketoglutarate oxidation (to 48·0 and 50·6 per cent of normal, respectively), while there was no change in thiamine-deprived and OTh-treated brains. The decreased rates of oxidative decarboxylation could be largely restored to normal by the *in vitro* addition of cocarboxylase. Brain and kidney homogenates did not oxidise α-ketoisovalerate and α-keto-β-methylvalerate.

70 Metabolic Pathways of Tetraiodothyro- and Triiodothyro-Acetic and Propionic Acids. E. V. Flock, J. L. Bollman and G. H. C. Stobie (U.S.A.).

Major metabolic pathways for thyroxine (T4) and 3:5:3′ triiodothyronine involve removal of an iodine atom from the benzene ring with the side chain and thus inactivation of these hormones. Conjugates of the products 3:3′:5′ triiodothyronine and 3:3′ diiodothyronine are excreted in the bile of dogs with livers but accumulate in the blood and urine of dogs without livers. Analogues of the thyroid hormones, labelled with ^{131}I in the 3′ or 5′ position, were studied in dogs with biliary fistulas and in dehepatized dogs. In dogs with biliary fistulas tetraiodothyroacetic acid (TETRAC) was metabolized much more slowly than T4; much less ^{131}I was excreted in bile or urine. Large amounts of unchanged TETRAC with smaller amounts of 3:3′:5′ TRIAC were found in the blood. Tetraiodothyropropionic acid (TETPROP) was partially deiodinated to 3:3′:5′ TRIPROP which accumulated in the blood as the amount of unchanged TETPROP decreased. Both of these compounds of propionic acid were excreted in bile as glucosiduronides. Triiodothyroacetic acid and 3:5:3′ triiodothyropropionic acid were rapidly cleared from the blood and excreted in the bile chiefly as glucosiduronides, with small amounts of the sulphoconjugates of 3:3′ diiodothyroacetic acid and 3:3′ diiodothyropropionic acid. In dogs without livers, the sulphoconjugates of these 3:3′ diiodo- compounds were found in larger amounts in both blood and urine, but the major metabolites appeared to be sulphoconjugates of 3′ monoiodo-derivatives.

71 Relations between Structure, Velocity of Biological N-hydroxylation and Toxicity of Aromatic Amines. H. Uehleke (Germany).

N-hydroxylation of aromatic amines in the body forms highly toxic methemoglobin forming hydroxylamine derivatives and seems to be important in allergy and carcinogenesis. Liver microsomes in the presence of TPNH and oxygen can perform the reaction. This is the first time that enzymatic N-hydroxylation has been achieved *in vitro* and that one of the few toxication mechanisms has been elucidated biochemically. The velocity of N-oxydation *in vitro* and *in vivo* depends on the chemical structure of aromatic amines. Aniline, naphthylamine and 2-aminofluorene are slowly oxydized. P-substitution of aniline by electron-attracting groups increases the velocity of N-hydroxylation in the animal and *in vitro*. Compounds with ionizable *p*-substituents (-COOH,-SO_3H) are not measurably attacked, presumably because of fat insolubility. Mono-N-alkylanilines are hydroxylated faster than aniline, the alkyl being removed as aldehyde in the reaction. Levels of oxydized amines in the animal and velocity of N-hydroxylation by liver microsomes are compared with toxicity and methemoglobia forming capacity of the corresponding amines. The reaction of N-hydroxylation will be discussed in view of other known hydroxylation mechanisms of aromatic compounds. The toxication mechanism described gives us a better understanding of the toxicity and side effects of many drugs.

72 Mechanisms of Activation and Induction of Rat Liver Tryptophan Pyrrolase. O. Greengard and P. Feigelson (U.S.A.).

The parenteral administration of a number of agents cause an increased level of tryptophan pyrrolase activity in the liver of rats. These agents include hormones, drugs and the substrate of the enzyme, tryptophan. The elevation of enzyme activity can be as much as 10-fold.

Tryptophan pyrrolase is an unusual heme-enzyme in that in normal liver much of it exists in the free apo-protein form, the presence of which can be revealed by the addition of hematin to the assay mixture.[1] Increased activity can therefore result both from saturation wth cofactor or from increased apo-protein concentration. Methods have been developed to distinguish between the two mechanisms, and differentiation between the modes of action of the various inducing agents has been achieved. The administration of cortisone or reserpine results in the accumulation of the apo-enzyme form of tryptophan pyrrolase in the liver. However, upon