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PROBLEM	SOLUTION OR CONTRIBUTION
What is acute uranium poisoning?	Described in 7 species of animals, in detail in dog, rat, rabbit
What types (kinds(of uranium poisoning are found?	1. Massive -- kills within 24 hours, like acute heavy metal poisoning, e.g., mercury 2. Acute -- kills in 1 to 3 weeks, azotemic like mercury bichloride 3. Chronic -- persistent - ill health due to daily exposures
How much uranium is required to produce (1) massive, (2) acute and (3) chronic types of poisoning?	Results mostly on rats: 1. Massive by 10-30 mg. per kg. body weight 2. Acute by 3-6 mg. per kg. 3. Chronic by diets containing 0.1 to 20% of various uranium compounds
Can uranium compounds be classed into groups on the basis of toxicity?	Three groups: 1. Highly toxic, e.g. TF_6 2. Mid-toxic, e.g., TNO_3 3. Slightly toxic, e.g., T_3O_8 Factor between groups: 1. is 10 to 100 times as toxic as 3 2. is 2 to 10 times as toxic as 3
Can maximum allowable concentrations of various uranium dusts in air be recommended based on animal toxicity studies?	Recommended maximum allowable concentrations have been supported: 1. toxic compounds 2. slightly toxic compounds
How important is the skin as a route of toxic exposure?	Hundreds of rats, rabbits and guinea pigs have been exposed to known doses on the skin. Rabbits are highly susceptible; man seems most resistant.

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Can minute amounts of isotopic uranium be received from concentrated 'gunk' solutions of other elements?	Dr. Flagg perfected a method of chemical recovery of 99 plus per cent of such uranium, simply and cheaply.
Can all radium be recovered from slags of uranium process	Dr. Flagg coöperated in developing coprecipitation procedures that work.
Can analyses of urines of industrial personnel serve as an index of fluoride exposure?	Probably best index - hundreds of samples have shown only a few higher than recommended level.
Necessity of stated daily maximum intake of fluoride to avoid poisoning.	Fluorine conference in N.Y.C. Set 6 ppm as project allowable exposure per day.
Is calcium metabolism connected with uranium and fluorine poisoning?	Deposition and storage of uranium and fluorine in bone make this certain. Calcium conference in N.Y.C. attended by experts from all parts of country contributed symposium on Ca metabolism.
Is F_2 high pressure burn a heat burn or a fluoride burn?	By aid of engineering device, controlled burns showed F_2 burn is probably almost entirely a heat burn. This dictated therapeutic practice.
Can teeth analysis help in estimating uranium or fluorine exposure?	Uranium analyses both chemical and histological on animals show marked effects of uranium and fluorine exposure. Human extracted teeth show uranium deposition in uranium workers.
How important is oral administration of uranium?	Studies on thousands of rats and on several dogs show toxicity less by oral route than by lung, but entirely possible to get poisoning with toxic compounds.
What sensitive qualitative chemical tests are available for uranium in solution?	Dr. Elinor Ware studied scores of chemicals and found several which serve as sensitive qualitative or semi-quantitative tests for uranium.

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Removal of uranium compounds from workmen's clothing	Agents for efficient removal were discovered and the techniques put into industrial practice.
Demonstration of the tolerance to uranium compounds	By administration of repeated small doses of uranium nitrate and UCl_4 rats have been made tolerant to lethal doses.
	By administration of repeatedly larger doses, rats have been made tolerant to 2 to 3 times the lethal dose.
Study of urinary phenols in uranium poisoning	Doses of 1.25 and 2.5 mgs. of uranium nitrate per kg. caused significant changes in urinary phenol output in rats.
Effect of diet on mortality from uranium nitrate	Alkaline diets decreased and acid diets increased the mortality in uranium poisoning.
A study of the degree of unsaturation of the phospholipid fatty acids of the U-poisoned kidney	A statistically significant decrease in the degree of unsaturation of the phospholipid fatty acids of U-poisoned rat kidneys has been demonstrated.
A study of the degree of unsaturation of the phospholipid fatty acids of the blood of U-poisoned rats	A 20% increase occurs in the degree of unsaturation of the phospholipid fatty acids of the blood of U-poisoned rats.
Do fatty livers occur in U-poisoned animals?	Fatty livers are commonly present in U-poisoned rats.
Sex difference in toxicity of uranium compounds	Female rats are more susceptible to uranium nitrate and UO_2F_2 administered intraperitoneally.
Distribution and excretion of U_6 after a single injection	The U_6 content of rat tissues after a single injection has been determined. The bone has been found to be the principal site of storage.
	The excretion of U_6 has been followed. In the rat excretion occurs primarily through the urine; the major portion of the excreted U comes out within 24 hours.
Distribution and excretion of U_4 after a single injection	The U_4 content of rat tissues has been determined. U_4 is excreted more slowly and in smaller amounts than U_6. Significant quantities of U accumulate in the liver after U_4 administration.

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A study of cholesterol in U-poisoned rats	Changes in cholesterol content and distribution have been shown to occur in the whole animal and in the blood, kidney, liver and adrenals of rats poisoned by U nitrate.
Effect of various agents on U nitrate toxicity	Adrenal cortical hormones and tocopherols (Vitamin E) caused a delay in mortality from U nitrate.
Distribution of U following inhalation	Increasingly larger amounts of U are stored in bone with length of time of exposure.
Development of a highly sensitive method for the determination of U in biological materials.	The fluorimetric method for determination of U has been refined so that 0.0000002 gms. may be determined quantitatively in biological material.
Analysis of industrial urine samples	Urine samples from industrial workers exposed to U have been analyzed by the fluorimetric method.
Study of urinary and bone citric acid in U-poisoning	Urinary citric acid increases after U administration; bone citric acid remains unchanged.

PROBLEMS	RESULTS
Inhalation toxicity of uranium-dust	
a. Sixty 30-day studies	Information is now available for the first time on the relative acute toxicity of 12 uranium dusts inhaled from atmospheres by laboratory animals. Under the same conditions of exposure, toxicity varied widely and varied directly with the solubility of the material. A single 2-hour exposure to 20 mg/m^3 of soluble UO_2F_2-dust was lethal for certain species of animals while dusts of insoluble U_3O_8, UO_2 were practically non-injurious in repeated daily exposures for periods of 30 days. Toxicity was evaluated from a number of criteria, namely, weight response, symptoms, biochemical, physiological, hematological and pathological changes. The distribution of uranium in the bodies of the exposed animals was also determined.
b. One-year studies	Information is now being accumulated on 5 of the industrially most important compounds of uranium, U-nitrate, UF_6, UCl_4, UF_4 and UO_2, on the toxicity for animals resulting from 1-year exposures. At least 2 levels of concentration of each dust are being tested in a daily 2-shift basis. At the levels selected from the information gained from the above 30-day studies, only slight or no toxicity has been observed with any of the dusts in animals that have been exposed to date for periods approximating six months. The dust levels selected were similar in most instances, and were far above in others, to those experienced in industry.
Studies Relative to Uranium-dust Toxicity	
Biochemistry	
Sensitive indicators of uranium-poisoning - urinary amino-acid creatine ratio	This represents a sensitive, entirely new test, that indicates a disturbance of the kidney resulting from entrance of uranium into the body. Appearing in the urine, variations may be ascertained easily. Control data in humans are remarkably constant.
Vitamin C Metabolism	Injection of uranium compounds produces an increase of vitamin C excretion in the rat. Vitamin C deficiency decreases resistance of guinea pigs to the inhalation of uranium dusts.
Sugar Metabolism	Carbohydrate metabolism was slightly altered at high levels of exposure to uranium dust and hydrogen fluoride vapor, but was by no means reduced to the diabetic level.

PROBLEMS	RESULTS
Sodium Bicarbonate Therapy	Dietary bicarbonate decreased mortality in rats injected with uranium salts, but appeared to increase the susceptibility of rats exposed to a high level of uranium dust. The beneficial effect of the bicarbonate in injection experiments appears to be related to the relief of secondary symptoms such as acidosis and to increased excretion rather than to decrease the intensity of the primary kidney damage.
Histopathology	
Intra-cellular phosphatase staining method	Shown for the first time, the kidneys injured by uranium regenerated morphologically, but still remain devoid of phosphatase, an enzyme essential for the proper functioning of the kidney.
Routine Pathology (H and E Sections)	Routine pathological examinations revealed that the kidney alone is the primary site of damage of uranium.
Renal Biopsy	The procedure of taking a specimen of the kidney during different periods of exposure of animals to uranium dust has been adopted in order to follow the course of toxicity in the individual animal.
Physiology	
Liver-function tests	Blood coagulation tests - prothrombin clotting times, fibrinogen levels, bromsulfalein retention on animals exposed to uranium dusts are being used as a measure of involvement of the liver in uranium poisoning.
Renal function tests	Inulin, Diodrast and Chloride clearance determinations on dogs exposed to uranium dusts are being made in order to ascertain whether the physiological dysfunction from uranium may prove a more sensitive indicator than morphological change.
Sperm Motility	Motility and other related properties of sperms were measured from rats exposed for several months to various uranium dusts without measurable effect.
Sampling of uranium dust in air	A filter paper of a special grade in a simple holder has proved the most efficient and easiest method of sampling air containing uranium dust.
Size-Distribution of uranium dust	The use of a multiple stage cascade impactor for the separation of circulating uranium dust in air has provided an effective means of grading dust size preparatory to analysis.

PROBLEMS	RESULTS
Particle-Size of Dust	A new selenium-coating method using a high vacuum technique has been applied to uranium dust which allows particles of 0.1 microns (10^{-5} cm.) to be seen.
Electron Microscope for Size Measurement of Certain Uranium Dust	Certain dust generated from a vapor with particle-sizes below the limit of visibility by optical methods required the use of the electron microscope.
Respirator Testing - Protection of the Worker Against Uranium dust	Various respiratory protective devices were given certification of acceptability for use against 12 uranium dusts.
Uranium Analyses - (Quantitative Chemical Method)	A rapid, simple micro-method for the determination of uranium dusts sampled by the filter-paper method in air by ferrocyanide has been developed for routine use in the inhalation studies.
Fluorine Analyses - (Quantitative Chemical Method)	A rapid colorimetric method by direct titration for determining micro-quantities of fluoride-ion has been devised which allows a 6-fold increase in time-saving.
Lung Retention of Uranium dust	A novel device employing electronic equipment for the determination of how much uranium dust and of what particle-size is removed from an atmosphere by inhalation and the site of deposition has been developed. These problems have not been heretofore satisfactorily solved.
Inhalation Toxicity of Fluorine Gas	Exposure of animals to various concentrations of fluorine-vapor for periods up to 30 days showed that 0.8 mg/m^3 is a safe exposure level. This concentration represents but a minute quantity in the atmosphere.
Inhalation Toxicity of Hydrogen Fluoride	Exposure of animals to different concentrations of hydrogen fluoride vapor in daily repeated intervals for periods of 30 days showed that 8 mg/m^3 constituted a safe level of exposure.
Inhalation Toxicity of Oxygen Fluoride - A by-product of fluorine disposal	This vapor was found to be probably the most toxic substance known. 0.5 ppm was lethal after 14 hours' exposure to laboratory animals; 0.1 ppm, however, was safe for animals undergoing 30-day exposure to this material.
Approximate Toxicity Tests on New Materials of Industrial Importance	Facilities were made available for the repeated testing of toxicity of all types of materials suspected of being injurious to the workers. Reports were issued within from 24 hours to one week after receipt of the materials.

PROBLEMS	RESULTS
Acidosis	Blood CO_2 decreases markedly Blood lactate normal Urine lactate up Urine volatile organic acids up Urine NH_3 very high in rabbits
Uremia	High NPN, urea, creatine, uric acid, rest nitrogen in blood with decreased excretion of same Urinary creatine high, especially associated with high blood NPN
Glycogen synthesis from lactate	Abnormal decrease after uranium injury Normal in animals exposed by inhalation
Effect of diet on uranium poisoning	A diet of greens and vitamin B_1 supplement has no effect in rabbits
Phenol red test	Rate of removal from blood decreased from injection and inhalation of uranium

PROBLEMS	RESULTS
What is the cause of primary toxic action of uranium on the kidney?	A general mechanism based on a great amount of experimental evidence has been formulated which states that hexavalent uranium filters through the glomerulus as the bicarbonate complex, and then becomes attached to the tubular cell walls after decomposition of this complex owing to various causes. The combination of uranium with the cell walls starts off a chain of events which results in extensive tubular damage. This mechanism might be applied in part to the action of other heavy metals.
How is uranium transported in plasma?	Hexavalent uranium is transported chiefly in the form of complexes with bicarbonate and protein in plasma. Increase in bicarbonate content of the plasma can greatly increase the ratio of the uranium bound as bicarbonate complex to that bound as protein complex. Tetravalent uranium is probably transported in plasma, chiefly in the form of a complex with protein, although some may be present as an insoluble oxide. To enter the plasma at all, U_4 probably must be injected directly.
Can uranium compounds be oxidized or reduced in the body?	Hexavalent uranium is very stable in the body, and in all probability is not reduced, although partial reduction in bone has not been definitely excluded. Tetravalent uranium is fairly stable at least for several hours in the body, although slow oxidation to hexavalent uranium may occur at least in the plasma. Pentavalent uranium is unstable and could exist at the most in the body only to the extent of a fraction of a per cent of the total uranium present. Trivalent uranium is unstable and would immediately be oxidized if injected into the body.
To investigate the chemical reactions of uranium compounds with proteins and to compare these reactions with corresponding reactions involving other heavy metals.	The reaction of uranium compounds with a number of proteins has been extensively investigated, and the observed results have been compared with corresponding results for other heavy metals. Applications of this work have been made in setting up a novel method for the analytical separation of uranium from bone and soft tissue. In addition, possible methods involving the use of uranium compounds for the analytical separation of plasma albumin from plasma globulin,

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PROBLEMS	RESULTS
	and for the determination of the isoelectric points of proteins are being studied. The effect of hexavalent uranium on the electrophoretic mobilities of plasma protein components has been studied, and it has been found possible to reverse this effect by chemical means.
To investigate the chemistry of uranium in the presence of certain non-protein complexing agents of biochemical importance.	The existence of some important uranium complexes as anions in solution has been discovered by means of electrophoresis experiments. The relative tendencies of these complexes to dissociate to yield uncomplexed ions of uranium have been determined roughly, and the dissociation constants of the acetate complexes of uranium have been measured by polarographic methods. The very small amount of pentavalent uranium which is present in solutions containing hexavalent and tetravalent uranium has been measured by polarographic methods. Oxidation-reduction studies of uranium ions and complexes have been carried out by several methods, with the aim of establishing what chemical forms of uranium would be stable in the body. Much of this work was done by means of the polarograph.
To determine a sensitive and early test for the toxic action of uranium on the body.	A. It was found that in addition to the appearance of urinary protein and the enzyme phosphatase, the enzyme catalase rises in concentration in urine very sharply after treatment of animals with very small doses of uranium compounds. This test was applied to rats, cats, dogs, rabbits, and workers industrially exposed to uranium compounds (by inhalation). B. A portable machine was developed for the rapid estimation of urinary catalase, based on a gasometric method.
To determine the effect of uranium compounds on enzymes and enzyme systems of the body. (These systems constitute almost the entire catalytic machinery of the body.)	The effect of hexavalent and in some cases of tetravalent uranium compounds on a number of enzymes and enzyme systems both *in vitro* and *in vivo* has been determined, and in certain cases detailed comparisons have been made of the action of uranium compounds and other heavy metals on highly purified crystalline enzymes. This work aided in setting up a general mechanism for the action of uranium compounds on the kidney and led to certain conclusions as to the possibilities of therapy in uranium poisoning.

PROBLEMS	RESULTS
To perform the best available laboratory tests on persons exposed to uranium compounds by inhalation with the purpose of determining whether or not toxic effects could be detected.	Such experiments were performed on ten soldiers at Oak Ridge, which included kidney and liver function tests, and sensitive urinary tests as well as analysis of urine for uranium and fluoride. In addition clinical examinations of the soldiers were made by the medical staff. No changes definitely attributable to uranium poisoning were observed.
To devise a rapid method to estimate the percentage of oxidized uranium (U_6) in UF_4 samples used in dietary experiments.	A rapid and very satisfactory polarographic method was devised.
To devise a rapid and sensitive method for the determination of fluoride	A catalytic method was devised, employing the polarograph. This method has not yet been used extensively.
To devise a sensitive method for the analysis of uranium compounds in chamber air.	A polarographic method proved to be feasible, but owing to the fact that in most cases less sensitive methods were sufficient, the polarographic method has not been employed extenisvely.
To devise a rapid method for the determination of uranium in gunk solutions or other solutions containing U_{235}, which would not result in loss of active material.	A modified polarographic method apparently has solved the problem in a highly satisfactory manner.
To determine whether F_2 could exist as a molecule in moist air samples.	It was determined by calculation that F_2 can exist as a molecule in moist air.
Does plasma protein contribute to the urinary protein in uranium poisoning?	By the use of immuno-chemical technique, it was found that plasma albumin contributed extensively to the urinary protein in uranium poisoning in rabbits.

PROBLEMS	RESULTS
What happens to various renal clearances after poisoning with uranium?	Knowledge of the effects of uranyl salts on renal clearance, indicating in a general way the site of action of the metal on the nepliron. Development of new methods of assessing kidney function not themselves involving actual clearance determinations.
How does the kidney handle uranium presented to it by the blood?	Uranium clearances under various conditions indicate that the metal is filtered through the glomerulus in the form of a bicarbonate complex, and that this complex may break down in the proximal tubule in varying degrees depending on alterations in the internal environment of the body. The knowledge so gained allows us to say what is the best treatment to prevent deposition of uranium on the proximal tubule.
What is the localization of uranium within the	Radioautograph studies indicate that uranium is spottily distributed throughout the kidney, with a tendency not to be deposited in glomerular areas. This finding reinforces the information gained from the second problem above and gives a basis for the interpretation of certain of the toxicological data.
The production of catalasuria by various renopathic substances.	Knowledge of the relation between dose of material injected, urinary excretion of catalase thereafter and the renal pathology produced gives the basis for application of the catalase excretion test as an indicator of renal toxicity.

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PROBLEMS	RESULTS
Establishing low concentrations of uranium dust in air	Development of mechanisms which provide dust concentrations in the range of .05 to 20 mg. per cu. meter (Std. deviation = ± 20%).
Rapid, approximate method of determining concentration of uranium dust in air	Prediction of concentration from the alpha particle emission rate of sample deposited on filter paper.
Method of obtaining short-time F_2 skin burns	Mechanism to permit timed (0.1 seconds to 10 seconds) exposure to F_2 under 10-100 PSI pressure.
Controlling flow of TF_6 at low rates	Construction of controlled temperature vaporization system with provisions for blending to various concentrations.
Determination of concentration and toxicity of fluorinated hydrocarbons in air	Toxicity levels and gas mask efficiencies established using infra red analytical method.
Practibility of Chrysler pump in determining amounts of F_2 in air	Tests indicated that method in original form gave erratic results on F_2. Further testing with tumeric paper gave good results with BF_3.