TABLE I. Comparison of Thermoprecipitability at 56°C of Whole Pyroglobulin, Normal F2 and Their H and L Preparations.

Test solution	O.D. at 280 mμ		
	Supernatant	Precipitate	% in precipitate
33.3% SAS fraction of myeloma serum	3.6	41	92
Normal F2	45.2	1.06	2
H chain myeloma serum	6.2	12.3	66
L " " "	1.13	.82*	42
H " F2	16.6	1.15	5.9
L " "	1.28	5.75*	81

* Soluble at 100°C.

globulins are different from those of Bence-Jones proteins since the pyroglobulin precipitate formed by heating to 56°C does not dissolve at 100°C, a characteristic of Bence-Jones proteins. This characteristic has been demonstrated also for L chains from normal F2 immunoglobulins(6) and this property was observed in both the F2 L and myeloma L chains separated in the present study.

Up to 10 bands of L chains have been observed in L chains prepared from normal immunoglobulin G when the chains were studied by starch gel electrophoresis at pH 7-8 with urea and glycine(5). An L chain preparation from an individual myeloma protein would have the migration of one of the 10 L chain bands(5). Such a result was observed with the L chain from the myeloma protein presently described.

We propose that the thermoprecipitable protein described be categorized as a subclass of myeloma immunoglobulin G, termed G myeloma P (for pyroglobulin) and that the thermoprecipitable H chain be termed γ (pyroglobulin) using presently recommended nomenclature(7).

Summary. A G myeloma pyroglobulin was reduced, alkylated and separated into "heavy" and "light" polypeptide chains. The type of thermoprecipitability which characterized the pyroglobulin was localized to the "heavy" chain. It is suggested that this myeloma protein is a subclass which could be termed G myeloma P (pyroglobulin).

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Use of Peritoneal Dialysis in Experimental Amphetamine Poisoning. (30511)

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The amphetamines are important therapeutic agents which are used primarily for suppression of appetite, reduction of fatigue, and central nervous system stimulation. Because these drugs also create a state of exhilaration and euphoria, they have been abused through non-medical channels. Considerable attention has been focused on their widespread and indiscriminate use(1). Although they are considered to be relatively nontoxic, serious clinical manifestations of toxicity have been associated with overdosage(2). A recent report(3) that death occurred as a result of methamphetamine poisoning, indicated that

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peritoneal dialysis might be a useful therapeutic measure in treatment of amphetamine toxicity. The present study is concerned with the effectiveness of peritoneal dialysis in experimental amphetamine poisoning.

Methods. All experiments were conducted on unanesthetized healthy mongrel dogs with weights ranging from 13.3 to 30.0 kg. The animals were fasted for 48 hours preceding the experiment. To each animal was administered 40 mg per kilogram of amphetamine sulfate suspended in approximately 300 ml of water through a nasogastric tube. This dosage was selected because it is 3 standard deviations greater than the previously determined LD_{50} for orally administered amphetamine sulfate in dogs(4) and therefore was presumed to be a surely lethal dose. For verification, a control group of 10 dogs was given this dose and was carefully observed for manifestations of toxicity. Five of these animals were observed in their cages and the remaining 5 were placed on the restraining board mentioned below. None of the animals in the control group were treated.

Thirteen animals comprised the treatment group. Each of these animals was placed on a specially devised restraining board which permitted excellent and painless immobilization. None of the animals was premedicated or anesthetized except for local infiltration with 1% lidocaine for the insertion of a peritoneal dialysis catheter and a venous cutdown catheter. An Ampro gastric sump tube, found in these experiments to be superior to standard dialysis catheters for the rapid inflow and drainage of peritoneal dialysis fluid, was introduced into the peritoneal cavity through a No. 28 trochar and guided into the lateral peritoneal gutter. The proximal end of the catheter was attached to a peritoneal dialysis set (Peridial Saftiset, Cutter Laboratories, Berkeley, Calif.). Standard commercial solutions‡ for peritoneal dialysis were used after first being warmed to the animal's body temperature. Dialysis was begun within 5 minutes following administration of amphetamine. Two liters of dialyzing solution were used in each dialysis cycle with 4 meq KCl added to each liter. In 8 animals, the glucose concentration of the inflowing solution was 4.25% (one liter of 1.5% and one liter of 7%) and in the remaining 5 animals, it was 7%. Average inflow time was approximately 10 minutes. The fluid remained in the peritoneal cavity for 10 minutes and then was drained as rapidly as possible by gravity drainage. When output flow ceased to be active, the next dialysis cycle was begun. Twelve dialyses were performed in each experiment over a period of approximately 6 hours. Fluid balance was maintained with intravenously administered 5% dextrose in water. Body temperature was continually monitored by a Yellow Springs Thermister Probe inserted into the muscles of the thigh. At the conclusion of each experiment, the animal was returned to his cage and observed for a period of at least 7 days. Seven days was the criterion for survival in this study.

‡ Peridial 1.5 and 7.0%, Cutter Laboratories, Berkeley, Calif.

TABLE I. Temperature Change and Duration of Survival in Control Group (10 Animals).

Animal No.	Temperature change (°C)	Duration of survival (min)
1	+3.08	140
2	4.07	195
3	4.4	345
4	5.8	294
5	4.95	180
6	4.07	135
7	4.95	90
8	5.28	130
9	5.72	240
10	5.28	205
Avg	+4.78 ± .77	195 ± 76

Amphetamine levels were determined by the modified methyl orange technique(5) with interfering substances removed by the buffer wash as recommended by Keller and Ellenbogen(6). Standard curves were prepared by adding known amounts of racemic amphetamine to the dialyzing solution.

Results. Table I lists changes in body temperature and duration of survival in the control group. All of the animals in this group died, average survival time being 195 minutes (±76). Manifestations of toxicity which occurred in each animal included extreme hyperactivity, tachycardia, tachypnea, and hyperpyrexia. The mean temperature elevation for the group was 8.7°F (±1.4).

TABLE II. Results in 13 Animals Treated with Peritoneal Dialysis.

Animal No.	% of administered dose recovered in dialysis	Temp change (°C)	Signs of toxicity	Result
11	2.2	− .82	0	Lived
12	2.3	−1.1	0	"
13	1.2	−1.26	0	"
14	1.3	− .44	0	"
15	1.9	− .11	0	"
16	.9	+4.18	+	Died at 17 hr*
17	7.3	+3.08	+	Died at 4 hr*
18	.9	− .11	0	Lived
19	1.7	− .88	0	"
20	3.9	− .11	0	"
21	3.1	+1.15	0	"
22	1.9	− .16	0	Died at 54 hr*
23	1.9	+ .22	0	Died at 16 hr*

* Time after administration of drug.

The results in the 13 animals treated by peritoneal dialysis are shown in Table II. Eleven (85%) showed no signs of toxicity, and 9 (69%) survived the 7-day period of observation with no apparent residual effects from the drug. Two of the 4 deaths occurred in the only animals in this group to have significant elevation of body temperature and other signs of toxicity during the dialysis. The other 2 deaths occurred in animals that showed no manifestation of toxicity during dialysis or during the period of observation prior to death.

The amount of amphetamine base recovered in the dialysate ranged from 0.9% to 7.3% of the administered dose, the average being 2.3% (±1.7). There was no correlation between per cent of drug extracted and either survival or manifestations of toxicity. The use of hypertonic 7% glucose solution in animals #19-#23 did not significantly increase the volume of dialysate or the per cent of amphetamine recovered.

Discussion. Peritoneal dialysis has been proved to be an effective form of therapy in the treatment of a variety of exogenous poisonings(7). In addition to effectiveness, this procedure has the benefit of being inexpensive, safe, and convenient. It can be performed at any medical facility and does not require highly specialized training(8).

Results of the present study indicate that poisoning with amphetamine can be managed successfully with peritoneal dialysis. Sixty-nine per cent of animals treated with dialysis as the only therapeutic measure survived a lethal dose of amphetamine sulfate. Furthermore, using a 6-hour period of vigorous dialysis, 11 of 13 animals demonstrated no manifestations of drug toxicity.

The percentage extraction of amphetamine in the dialysate was extremely low (2.3 ± 1.7). This recovery was not sufficient to have significantly altered the toxicity or lethality of the drug. Observations have been reported previously in which the successful performance of dialysis in the treatment of exogenous poisonings has been associated with poor recovery of the toxic agent(9). It is possible that a metabolite or bound amphetamine compound was actively extracted which was not measured by the assay techniques employed. It thus appears that recovery in the dialysate should not be a criterion for the efficacy of peritoneal dialysis in management of amphetamine poisoning. Favorable results may be obtained in spite of apparent low extraction of the drug.

Summary. Each of 10 unanesthetized dogs, constituting a control group was administered 40 mg per kilogram of amphetamine sulfate orally. Each animal showed signs of drug toxicity and all died within 6 hours. An experimental group of 13 dogs was given a similar dose and was subsequently treated with peritoneal dialysis. Eighty-five per cent of the treated group showed no signs of toxicity and 69% survived for at least 7 days without residual effects from the drug. The quantity of amphetamine recovered in the dialysate was negligible in spite of the favorable results. It is suggested that a metabolite or bound amphetamine compound was removed that was not measured by standard assay techniques. Peritoneal dialysis would appear to be an effective form of therapy for amphetamine poisoning.

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Effect of Inhibition of Oxidative Phosphorylation on Incorporation of Inorganic Phosphate (P^{32}) into Liver Mitochondrial ATP and Phospholipids.* (30512)

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A number of enzymes found in mitochondria involved in oxidative phosphorylation contain phospholipids(1,2). However, if the phospholipids are removed there is a decrease in enzymatic activity and activity is restored with the addition of phospholipids(3). Compounds such as bilirubin(4), dinitrophenol (5), arsenate(6,7), and oligomycin(8,9) have been shown to inhibit oxidative phosphorylation. All of these compounds inhibit the incorporation of inorganic P (P_i^{32}) into the individual liver mitochondrial phospholipids (10). Heart mitochondria isolated from rats fed a magnesium deficient diet for 4 to 8 days exhibit uncoupling of oxidative phosphorylation with a decrease in P/O ratios (11) and result in decrease synthesis of the individual phospholipids of heart mitochondria(12). However, in none of the above conditions was the synthesis of ATP investigated. With these observations in mind, experiments were undertaken to determine the effect of inhibition of oxidative phosphorylation on incorporation of P_i^{32} into liver mitochondrial ATP and phospholipids.

Methods. Male albino rats of Sprague-Dawley strain, weighing 110-150 g, were divided into groups. The animals in Group 1 were injected intraperitoneally daily with dinitrophenol (30 mg/kg for 3 days), in Group 2, rats were injected with a single dose of arsenate (14 mg/kg). Control rats for each group received intraperitoneal injections of saline. Intraperitoneal injections of 200 μc of P^{32} as NaH_2PO_4 were administered one hour following injection of the above compounds. One hour after P_i^{32} administration the animals were killed by decapitation. This time interval corresponds to the ascending part of the specific activity time curve, where synthesis of ATP occurs to a degree greater than breakdown(13).

The liver was removed, rapidly weighed, homogenized in a Potter-Elvehjem tissue grinder with teflon pestle in ice-cold 0.25 M sucrose containing 0.00018 M $CaCl_2$. The homogenate was divided into 2 fractions. From one fraction acid-soluble phosphorus was removed by extracting with 10% TCA, and the radioactivity(14) and acid soluble phosphorus(15) were determined. The other homogenate fraction was separated into cellular fractions by the method of Hogeboom *et al*(16). The homogenate was centrifuged at 600 $\times$ *g* for 15 minutes to remove the nuclei. The mitochondria were separated from the supernatant by centrifugation at 10,000 $\times$ *g* for 15 minutes. The mitochondria were washed twice with 0.25 M sucrose and recentrifuged at 10,000 $\times$ *g* for 15 minutes

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