was reduced and treated as described above (yield, 70 mg., 74%).

The free base, m.p. 160° [lit. m.p. (1) 160°], and the salicylate, m.p. 223–225° [lit. m.p. (4) 223.5–224.5°], prepared from VI hydrochloride showed no melting point depression when mixed with authentic samples. The IR spectra of the hydrochloride and the salicylate were identical with authentic samples.

Pellotine (VII) Hydrochloride—*Method A*—Sodium borohydride (20 mg.) was added in small portions over a period of 0.5 hr. to a stirred solution containing VI (38 mg.) and formaldehyde (2 drops, 37%) in methanol (5 ml.) at room temperature. The solution was then added to water (5 ml.) and extracted with chloroform (3 × 10 ml.). The chloroform extract was dried over anhydrous sodium sulfate and then evaporated to dryness. The residue was dissolved in a small quantity of methanol–chloroform–ether, and dry HCl gas was passed through the solution. Colorless needles crystallized (yield 35 mg., 75%).

Method B—A mixture of V (90 mg.), 6 *N* HCl (20 ml.), and palladium-on-charcoal (200 mg., 5%) was reduced at atmospheric pressure and room temperature for approximately 24 hr. The mixture was filtered and the filtrate concentrated *in vacuo* at <50° using a film-flash evaporator. Ethanol–benzene was added from time to time to azeotrope the water. The solid residue was crystallized from methanol–ether (yield 87 mg., 89%).

The free base, m.p. 116° [lit. m.p. (1) 111–112°], showed no melting point depression when mixed with authentic pellotine. The IR spectra of the base and hydrochloride were identical with that of authentic samples.

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Pentaerythritol Tetranitrate Sustained-Release Tablets: Relation of *In Vitro* Release of the Drug to Blood Pressure Changes after Administration to Anesthetized Cats

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Abstract □ The *in vitro* release of pentaerythritol tetranitrate (PET) from a sustained-action PET tablet was determined using USP tablet disintegration apparatus with the modification that 100-mesh stainless steel wire cloth replaced the usual 10-mesh. The immersion medium was simulated gastric fluid for the first 2 hr. and simulated intestinal fluid for the subsequent 10 hr. There was a gradual release of the drug and 80 to 100% of the drug was dissoluted at the end of 12 hr. The tablet when administered by mouth to anesthetized cats produced a 28 to 30% fall in arterial blood pressure which persisted for 12 hr. A good correlation between the rate of release of the drug *in vitro* and the *in vivo* effect in sustained lowering of the blood pressure was obtained. The *in vitro* method described is proposed for the quality control of sustained-action PET tablets.

Keyphrases □ Pentaerythritol tetranitrate sustained-release tablets—drug release □ Blood pressure changes—pentaerythritol tetranitrate release □ Dissolution rates—pentaerythritol tetranitrate tablets □ *In vivo-in vitro* release rates correlation—pentaerythritol tetranitrate tablets

Difficulties were experienced in testing the quality of sustained-release tablets of pentaerythritol tetranitrate using procedures usually adopted for the quality control of sustained-release preparations. In the dissolution test the concentration of the drug released in the bath fluid from the tubes containing tablets is estimated at various intervals. In the assay of pentaerythritol tetranitrate such a procedure could not be adopted as the analysis is based on the estimation of nitrates. To estimate nitrates in the bath fluid, large volumes of the solution had to be evaporated to dryness before extraction with glacial acetic acid (1). In the *in vivo* test the determination of the rate of absorption of the drug would involve the collection of large volumes of blood at frequent intervals and estimation of the drug in these blood samples. Both procedures were impractical.

In the dissolution test, if the remnants of the tablets in the disintegration apparatus and not the bath fluid were collected at different intervals, the estimation of pentaerythritol tetranitrate would be possible and the *in vitro* rate of release of the drug from the tablet could be calculated. The drug lowered blood pressure and a sustained fall in blood pressure could be observed when the sustained-release tablet was administered to cats. A study, therefore, was undertaken to establish a relation, if any, between the *in vitro* release of the drug and the lowering of blood pressure in cats after administration of sustained-release pentaerythritol tetranitrate tablets. A good correlation was observed between the *in vitro* release of the drug and the fall in

Table I—Percent Release of Pentaerythritol Tetranitrate from a Sustained-Release Tablet Containing 80 mg. of the Drug[a]

1 hr.[b]	2 hr.[b]	4 hr.[c]	6 hr.[c]	8 hr.[c]	10 hr.[c]	12 hr.[c]
16 ± 1.5 (9)[d]	16 ± 1.5 (9)	32 ± 3.5 (9)	45 ± 3.7 (8)	56 ± 2.8 (9)	69 ± 2.9 (8)	90 ± 2.0 (9)

[a] Values are mean ± standard error. [b] Simulated gastric fluid. [c] Simulated intestinal fluid. [d] Figures in parentheses indicate the number of observations.

Table II—Relation of *In Vitro* Release of Pentaerythritol Tetranitrate (PET) to Percent Fall of Blood Pressure in Cats after Administration of a Sustained-Release Tablet

	2 hr. (10)[a]	4 hr. (10)	6 hr. (10)	8 hr. (10)	10 hr. (5)	12 hr. (5)
% fall of blood pressure[b] from basal in cats after feeding sustained-release PET tablet, 15 mg./kg.	29 ± 3.0	26 ± 2.1	31 ± 3.8	32 ± 3.0	25 ± 2.1	19 ± 3.8
Release of PET (mg.)[b] *in vitro* from a sustained-release tablet containing 80 mg. drug	14 ± 1.1	10 ± 1.4	10 ± 1.2	10 ± 1.6	14 ± 3.0	11 ± 2.9
Coefficient of correlation	0.887	0.919	0.926	0.980	0.801	0.985

[a] Figures in parentheses indicate the number of samples used. [b] Values are mean ± standard error.

blood pressure which indicates that the *in vitro* procedure described in this communication can be used for the quality control of the sustained-release dosage form of pentaerythritol tetranitrate.

EXPERIMENTAL

Dissolution Rate Determination—The sustained-action pentaerythritol tetranitrate (PET) tablets under study were two-layered compressed tablets of which one layer contained 10 mg. PET for immediate release and the second layer contained 70 mg. of the drug for sustained release.

For the determination of the dissolution rate, the tablet disintegration apparatus as described in USP XVII (2) was used with the modification that 100-mesh stainless steel wire cloth was attached to the under surface of the lower end of the disintegration tubes in place of 10-mesh usually used for determination of disintegration of tablets. One tablet was placed in each of the tubes and the machine was switched on using simulated gastric fluid with a pH of 1.2 (3). After 2 hr. the machine was switched off. One of the tubes used for disintegration was withdrawn and the remnant of the tablet transferred into a 150-ml. conical flask. The simulated gastric fluid was replaced by simulated intestinal fluid (4) adjusted to pH 7.5. The machine was switched on again and operated for another 10 hr. At intervals of 1 hr., one of the disintegration tubes was withdrawn and the remnant of the tablet was transferred into a 150-ml. flask. The remnants of the tablets were extracted with glacial acetic acid on a boiling water bath, suitably diluted with the acid to bring to a definite volume, and PET content was estimated (1). Twenty of the tablets were crushed to powder and PET content was estimated in an aliquot to obtain the contents in one tablet before disintegration. From the contents of PET in the remnants of tablets after various hours of dissolution, the rate of release of the drug from a tablet was calculated. The results are given in Table I.

Blood Pressure Studies—Cats weighing 2.5–3 kg. were anesthetized by intramuscular injection of sodium phenobarbital, 150 mg./kg., dissolved in 4 ml. of water for injection. After the animal was under the anesthesia the left common carotid artery was cannulated in the usual way to record the arterial blood pressure on a kymograph. The trachea was connected to a recording tambour for the recording of respiration. When the initial blood pressure and respiration were recorded the sustained-release PET tablet, 15 mg./kg., was fed to the animal through a stomach tube, 10 ml. water was injected through the tube so as to push the tablet into the stomach, and blood pressures were recorded at intervals. Fifteen milligrams PET per kilogram body weight was found to produce a marked fall in blood pressure without showing any undesirable effect. The relation of percent fall of blood pressure to *in vitro* release of PET with statistical analysis is shown in Table II.

When quick-release PET was administered in the same dose, there was a 40% fall in blood pressure which came to the initial level within 6 hr. The anesthesia itself did not produce any marked change in the blood pressure.

RESULTS AND DISCUSSION

The results in Table I indicate that the *in vitro* release of PET was gradual and 90% of the drug was released in the course of 12 hr. The results were found reproducible, within the limits of the assay methods, from lot to lot of the same batch and in different batches. Table II shows that the percent fall in blood pressure, after administration of PET, at different hours had a good correlation with the release of PET in the *in vitro* experiment.

With regards to PET, controlled studies in humans based on the blood concentration of the drug or its degradation product are not possible as the rate of release of the drug is only 5 to 7 mg./hr. and concentration of the drug in the blood is too low to be analyzed. The *in vitro* test, therefore, may be used as a quality control procedure for the evaluation of a sustained-release preparation of pentaerythritol tetranitrate tablets.

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