DRUG DEVELOPMENT AND INDUSTRIAL PHARMACY, 11(4), 815-831 (1985)

IN-VITRO DISSOLUTION PROFILE AND OPTIMIZATION OF ASSAY TECHNIQUE FOR SUSTAINED RELEASE VITAMIN C PRODUCTS

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ABSTRACT

Six commercial sustained release Vitamin C products in the form of encapsulated coated pellets and compressed matrices were evaluated. A modified NF XIV procedure was used to monitor the release rate under two separate conditions. One condition involved bubbling of nitrogen gas in the extracting fluid in the bottle and displacement of air with nitrogen in the head space. The other involved no deaeration of extracting fluid with nitrogen and no displacement of air in the head space. Recovery of Vitamin C from some of the products evaluated was significantly higher, especially at higher pH of extracting fluid (pH > 4.5) using

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nitrogen gas. Release profiles varied significantly among the products under the conditions investigated.

INTRODUCTION

In recent years, the study of dissolution of drugs from solid dosage forms has become increasingly important. The rate and extent of dissolution from tablets, capsules and pellets affect both the absorption and therapeutic effect of a drug. Different formulations of the same drug may exhibit different absorption characteristics and, subsequently, different therapeutic activity. (1)

In absence of any compendial dissolution guidelines for sustained release products, there is disagreement as to the apparatus and the method that should be used as standard. Many dissolution techniques have been conceived, improved and occasionally replaced. A simple inexpensive apparatus and method that could be used for most products would be ideal. The rotating bottle method (2) is one method which involves inexpensive apparatus. However, it is gradually moving into extinction and all new sustained release dosage forms are being tested by conventional, compendial dissolution methods, primarily because the rotating bottle method does not lend itself to automated testing.

Numerous factors influence dissolution testing, dissolved gas being one of them. The dissolved gas may significantly change the pH of

media for e.g. distilled water with dissolved air may have a pH as low as 6.0 compared with a pH of 7.2 for completely deaerated distilled water. The rotating bottle method provides ease of not only displacing dissolved gases in the extracting fluid but also of replacing the air head above the medium in the dissolution vessel. Since all liquids are in equilibrium with surrounding gas at the gas/liquid interface and at any given pressure and temperature a portion of the gas is dissolved in the liquids in the dissolution process, it may interfere and change significantly the nature of the active ingredient and alter analytical valves.(3)

The objective of this study was to determine the in vitro release characteristics of some commercial sustained release Vitamin C products under similar conditions of dissolution testing and evaluate the product performance in vitro to support its label claim for sustained release. The effect of deaeration of the extracting fluid and displacement of the air in the head space of dissolution vessel with inert nitrogen gas on the assay of ascorbic acid by dichlorophenolindophenol titration was also investigated.

EXPERIMENTAL

Materials

The following materials were used: Metaphosphoric acid[1], glacial acetic acid[2], 2,6-dichlorophenol-indophenol sodium[3], sodium

1-8 Fisher Scientific, Medford, MA.

bicarbonate[4], sodium hydroxide[5], hydrochloric acid[6], monobasic potassium phosphate[7], sodium chloride[8], Ascorbic Acid USP[9], and Whatman no. 1 filter paper.[10] All chemicals, except Ascorbic Acid, were reagent grade and were used as received.

Six commercial sustained release Vitamin C dosage forms tested were purchased from the market and are described in Table I.

Extracting Fluids

Gastric fluid (pH 1.2) and intestinal fluid (pH 7.5) were prepared according to the method described in USP XX (4) without the addition of enzymes. The other test fluids, pH 2.5, 4.5, and 7.0 were made using a mixture of gastric fluid and intestinal fluid in an appropriate ratio to give the desired pH within $\pm$ 0.05 units.

Release Rate

One sustained release tablet/capsule was transferred to each of the six dissolution bottles and 60 ml of extracting fluid, preheated to 37°C, was added to each bottle. Nitrogen gas was then bubbled into the solution and in the bottle for 5 minutes in order to displace air in bottle

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9 Hoffman LaRoche, Nutley, N.J.

10 W & R Balston, Ltd., England

TABLE I

Description of the 500 mg Vitamin C sustained release dosage forms examined.

Product Name & Designation	Description	Claim
A. Ultra Cee[1]	Encapsulated coated pellets	8 hour timed release
B. SNR Vitamin C[2]	Compressed matrices	Timed release
C. CVS Vitamin C[3]	Encapsulated coated pellets	Timed release
D. Vitamin C[4]	Encapsulated coated pellets	Timed release
E. C-Time 500[5]	Compressed matrices	Natural long-acting
F. Vitamin C 509 formula[6]	Compressed matrices	Timed release

[1]Hudson Pharmaceutical Corp., N.J.

[2]Nature-made nutritional products, CA.

[3]Consumer Value Stores, R.I.

[4]Rexall Corporation, MI.

[5]Nature's Bounty, Inc., N.Y.

[6]Food Plus, Inc., N.J.

head space. The bottles were then capped tightly and rotated for a predetermined time period at 40 ± 2 r.p.m. in water bath maintained at 37 ± 0.5°C using the rotating bottle apparatus.[1]

At the end of the time period all the bottles were removed and the tablet/beads residue allowed to settle. The supernatant liquid was decanted and filtered through Whatman no. 1 filter paper and the filtrate was retained for analysis. 60 ml of the extracting fluid was again added to each of the 6 bottles and the procedure repeated. The pH of the extracting fluid and the time allowed for rotation with each fluid along with sampling interval were as follows:

pH of extracting fluid	Time allowed for extraction	Sampling interval
1.2	1 hr	1 hr
2.5	1 hr	2 hr
4.5	1 hr	3 hr
7.0	2 hr	5 hr
7.5	2 hr	7 hr

At the end of 7 hours the residue was retained for analysis.

A similar procedure, with the omission of nitrogen, was employed as control for each of the products tested.

[1]E.D. Menold, Lester, PA.

Analytical Procedure

Assay of ascorbic acid was done according to USP XX (4) procedure.

A known quantity of filtrate from each sample was transferred to a 100 ml volumetric flask. The residue remaining after 7 hours was crushed and transferred to a 100 ml volumetric flask. 25 ml of metaphosphoric acetic acid T.S. (4) was then added to each and volume made up with distilled water. 4 ml of this solution was pipetted into a conical flask along with 5 ml of metaphosphoric acetic acid T.S. and titrated against standard dichlorophenol indophenol solution. The ascorbic acid content in each sample was determined and subsequently the amount of ascorbic acid released or recovered from each of the six samples at various sampling intervals was calculated. Necessary correction for the volume of standard dichlorophenol indophenol solution consumed by a mixture of metaphosphoric acetic acid T.S. and water was made.

RESULTS

Dissolution Profile

Of the six products evaluated only one product, Product A, claimed a definite 8 hour timed release profile on the label. Products

A, B, C, and D all showed a gradual release with 35-45% of the labelled amount being released in the 1st hour, the remaining being released over a 7 hour period. Product E and F gave a release profile quite different from the rest of the products. Product E, claiming long-acting, released about 27% of the labelled amount in the first hour under gastric pH conditions (pH 1.2) and nearly 100% of the labelled amount was released at the end of 3 hour period. Also the total amount of ascorbic acid recovered from each of the tablets of Products E calculated from cumulative release and residue analysis, was found to be an average of 610 mg, 22% in excess of its labelled amount. In the case of Product F, 61% of the drug was released in the first hour and 85% in the second hour of dissolution testing thereby providing little support to its claim for timed release.

None of the products tested exhibited pseudo-zero order release rates. However, when the total amount released was plotted versus the square root of time, Products A, B, and C showed a release rate that was linear and appeared to follow the theoretical relationship proposed for solid drugs dispersed in solid matrices (5).

Ascorbic Acid Assay and Recovery

The percentage of ascorbic acid recovered with respect to the label claim (500 mg) under 2 sets of dissolution conditions, with and without nitrogen, for all the products evaluated are listed in Tables II

TABLE II

Cumulative percentage of labelled amount of Vitamin C released against time. (Average of six samples).

Product A, B, and C.

Time in Hrs.	Product A				Product B				Product C			
	without nitrogen		with nitrogen		without nitrogen		with nitrogen		without nitrogen		with nitrogen	
	%	CV	%	CV	%	CV	%	CV	%	CV	%	CV
1	42.46	4.29	46.84	4.10	35.81	3.51	39.41	2.01	42.18	3.56	43.20	4.88
2	57.01	2.89	63.10	3.37	51.07	3.61	57.06	2.34	61.75	4.19	62.91	4.02
3	65.78	2.29	72.33	2.85	63.26	2.93	69.82	1.93	72.20	4.11	73.46	3.54
5	73.02	2.27	82.24	3.15	72.12	3.07	81.71	1.57	81.60	4.63	88.58	3.92
7	89.92	1.72	99.11	2.11	77.48	2.73	88.18	2.04	93.26	4.20	101.57	1.33
Res*	97.32	1.73	106.77	1.46	89.36	3.30	100.87	2.63	101.51	3.06	104.08	1.18

*Residue

TABLE III

Cumulative percentage of labelled amount of Vitamin C released against time (Average of six samples).

Product D, E, and F.

Time in Hrs.	Product D				Product E				Product F			
	without nitrogen		with nitrogen		without nitrogen		with nitrogen		without nitrogen		with nitrogen	
	%	CV	%	CV	%	CV	%	CV	%	CV	%	CV
1	34.79	14.74	36.34	8.93	25.87	13.81	27.18	14.05	55.05	3.41	61.72	4.41
2	60.66	9.40	63.33	4.95	61.86	6.98	70.63	10.87	74.32	2.99	85.24	3.52
3	76.59	7.80	77.36	3.42	96.19	3.19	103.22	7.00	84.04	2.62	96.39	3.52
5	84.07	4.28	85.59	3.06	109.36	2.01	120.95	6.67	91.01	2.23	104.00	3.71
7	89.63	3.57	92.33	2.54	109.62	1.92	121.84	6.66	91.59	2.19	105.04	3.86
Res*	98.62	4.31	101.82	2.31	109.68	1.92	122.10	6.64	91.74	2.21	105.67	3.84

*Residue

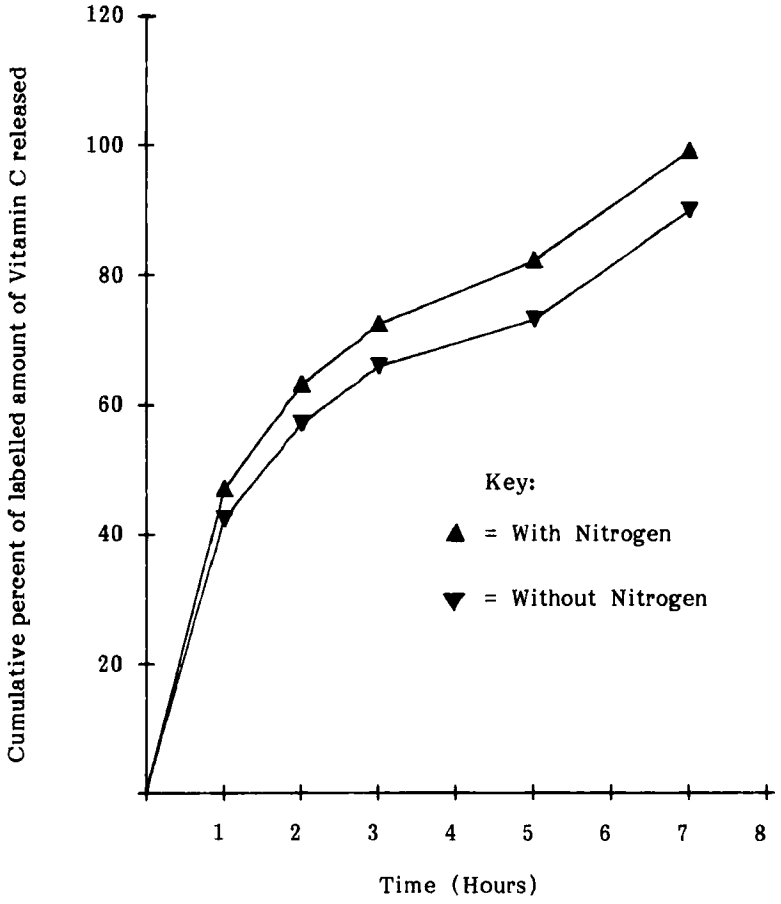

FIGURE I

Release Profile of Product A

and III. The release profiles for Products A through F are shown in Fig. I-VI.

Recovery of ascorbic acid by 2,6-dichlorophenolindophenol assay in case of five out of six products evaluated was significantly higher when nitrogen was used for deaeration. With the exception of Product D, all products evaluated showed substantial difference between the

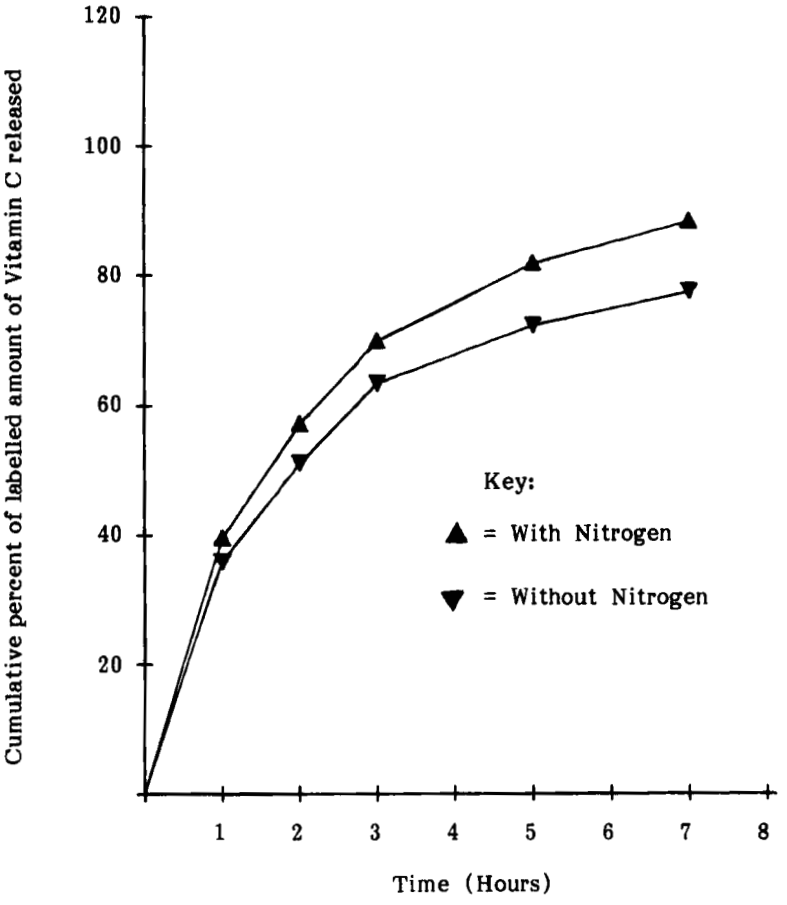

FIGURE II

Release Profile of Product B

mgms of ascorbic acid released with and without nitrogen, especially at higher pH of extracting fluid (pH 7.0 and 7.5) and for longer sampling intervals. The total amount of ascorbic acid recovered during dissolution runs was approximately 50 mgms more when nitrogen was used as opposed to control runs, without nitrogen, for Products A, B, E, and F.

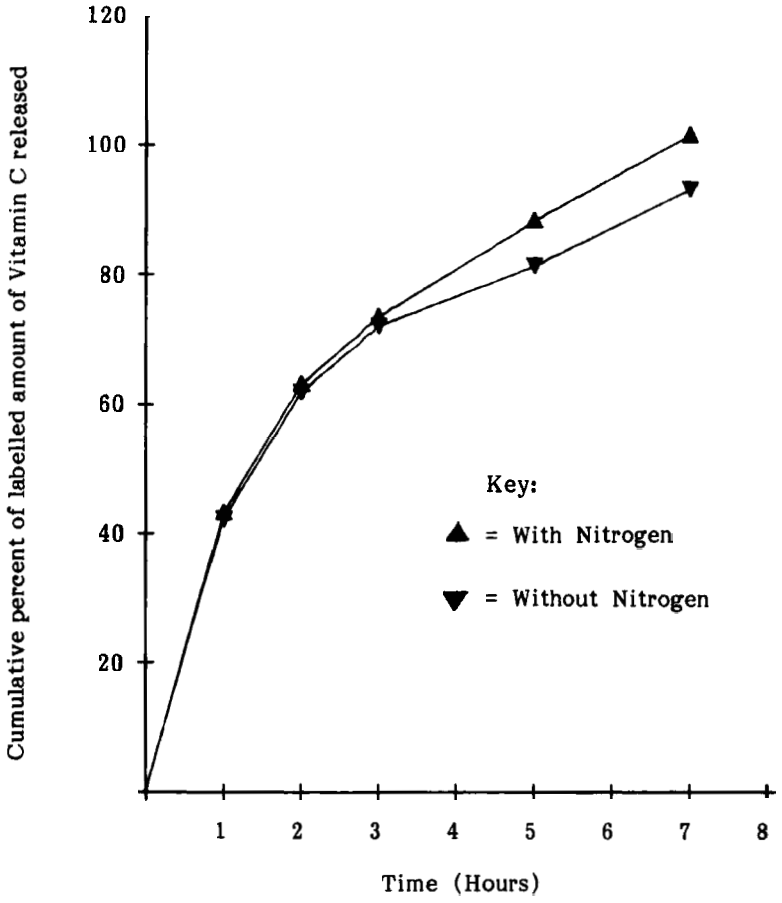

FIGURE III

Release Profile of Product C

DISCUSSION AND CONCLUSIONS

The rationale for administration of sustained release Vitamin C dosage forms is justified in cases of deficiency since Vitamin C is not stored in the body to any significant extent and a daily intake is required to avoid deficiency. A study by E. Stewart Allen (6) revealed that the mode of administration of ascorbic acid had a marked effect on

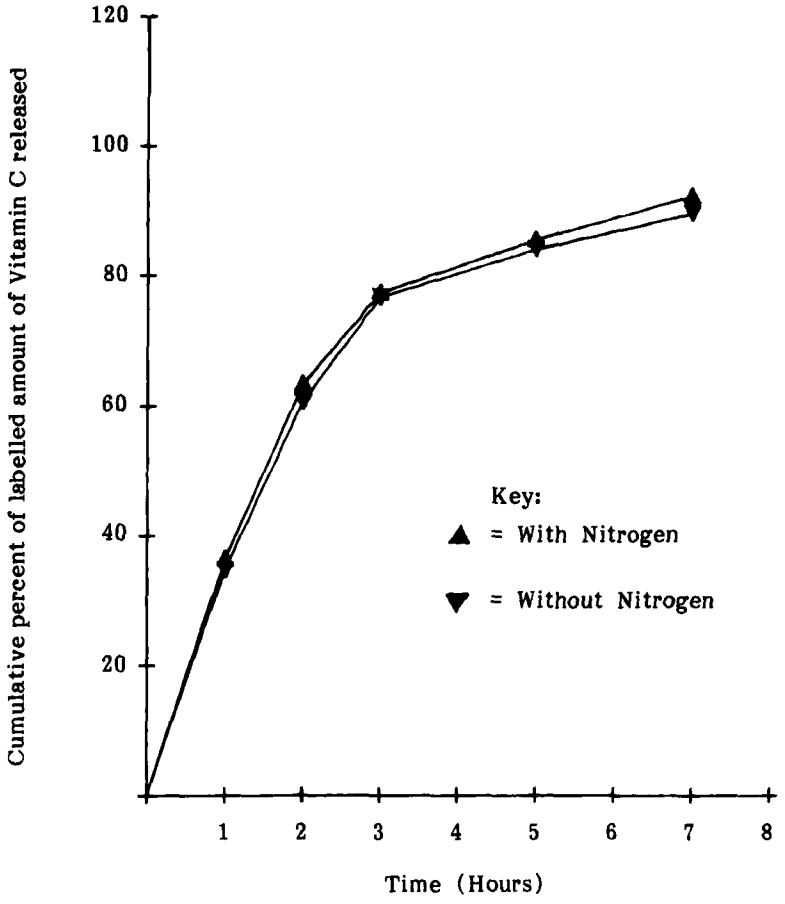

FIGURE IV

Release Profile of Product D

the bioavailability of the vitamin for metabolic utilization by the body, particularly when dosage levels are indicated.

This comparative study of the release of Vitamin C in vitro, from different sustained release dosage forms, showed that the rate of dissolution or release differed greatly between some products, and this

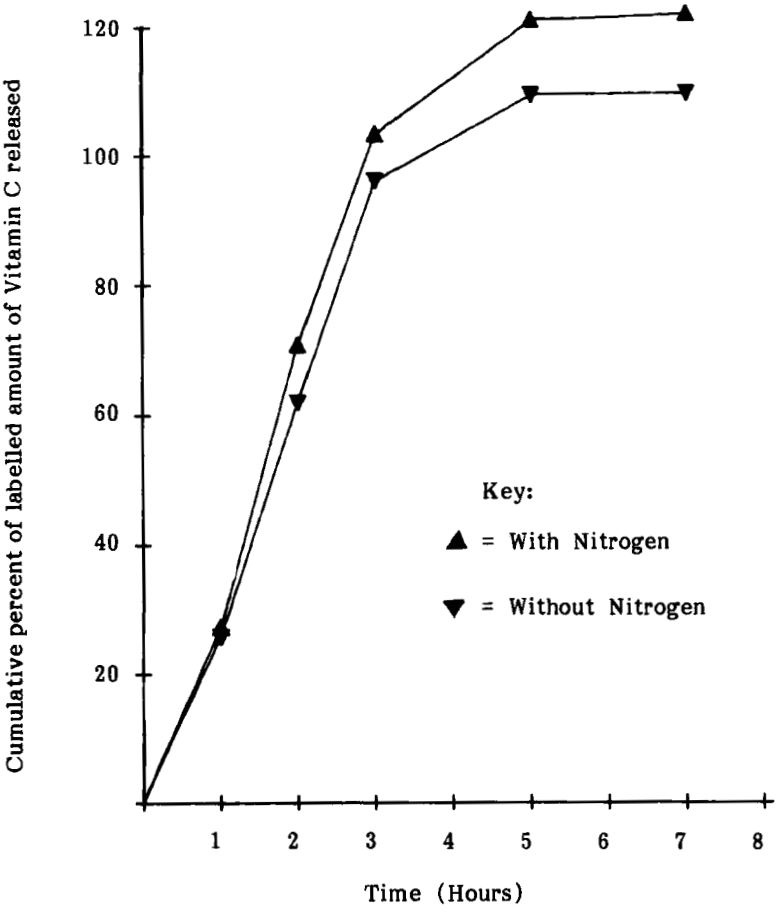

FIGURE V

Release Profile of Product E

would probably be reflective of their bioavailability. It is clear that only a comparable study in vivo would provide confirmation of this hypothesis which is based on examination of the kinetics of release in vitro. However, the primodial and well established influence of the dissolution rate of drug from its sustained release dosage form on the absorption indicates that in absence of comparative pharmacokinetic

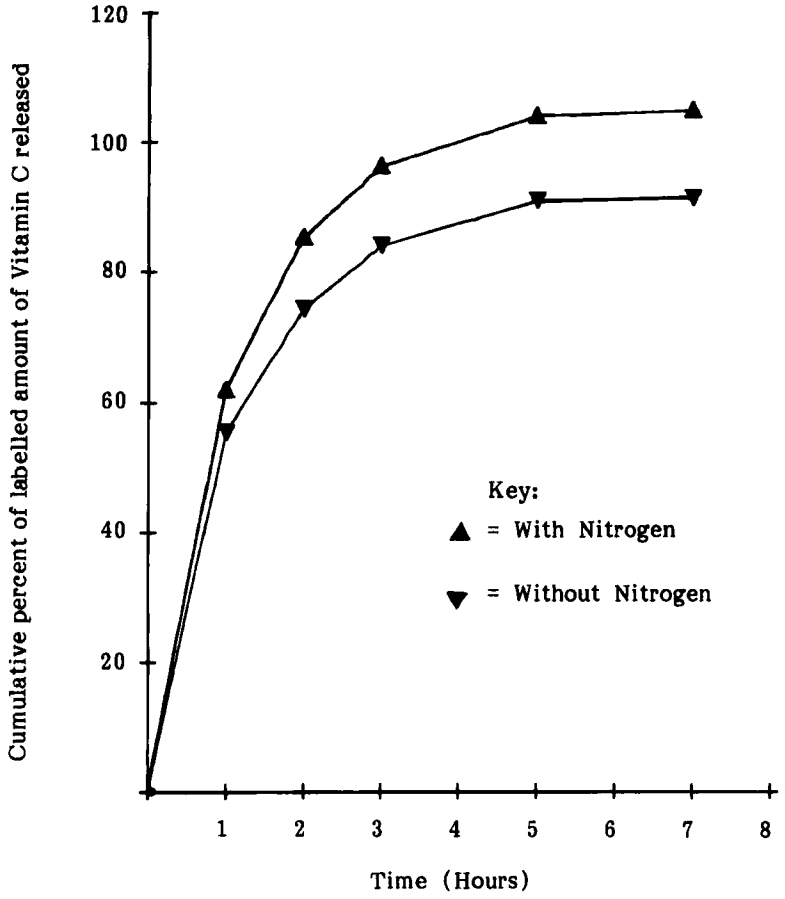

FIGURE VI

Release Profile of Product F

studies in man, the in vitro release profiles of the commercially available products can be a useful guide in determining their adequacy to support the claim for sustained release, despite its limitations.

Loss of ascorbic acid was found during the assay of dissolution samples obtained without using nitrogen, apparently due to oxidation of the drug during the extended period required for dissolution profile at

higher pH of extraction fluid. L-ascorbic acid, like all -keto-ene-diols, is a powerful reducing agent in acid and neutral solutions. It reacts with dichlorophenolindophenol and most methods for determination of the vitamin are based upon its reducing properties. (7) Aqueous solutions of L-ascorbic acid undergo oxidation, sluggishly but reversibly, under mildly acidic conditions when exposed to air to form dehydroascorbic acid. It breaks down rapidly and irreversibly in alkaline solution to form decomposition products including diketogluconic acid. (8) The oxidation was eliminated by replacing oxygen/air with nitrogen by degassing the various pH extraction fluids and using a nitrogen head.

REFERENCES

1. R.J. Timko and N.G. Lordi: J. Pharm. Sci., 67: 496 (1978).

2. "The National Formulary," 14th ed., Mack, Easton, PA (1975).

3. W.A. Hanson: "Handbook of Dissolution Testing," Pharm. Tech., Oregon, (1982).

4. "The United States Pharmacopeia," 20th rev., Mack, Easton, PA (1980).

5. T. Higuchi: J. Pharm. Sci., 52: 1145 (1963).

6. E.S. Allen: Curr. Thera. Res., 11: 745 (1969).

7. "Vitamin C-Merck Service Bulletin," Merck and Co., New Jersey (1956).

8. A.N. Matrin, J. Swarbick, A. Cammarata: "Physical Pharmacy," Lea and Febiger, Philadelphia (1969).