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Letter to the Editor

Lack of Degeneration of Penicillin in Urine during Storage of Specimens for up to Twelve Weeks

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The microbiological disk agar-diffusion method is widely used for the assay of penicillin concentrations in body fluids such as serum and urine. Specimens have to be assayed in batches for cost-effectiveness. This necessitates storage of specimens for various periods of time.

Penicillin is known to degenerate in stored specimens, even if they are frozen (Sutherland & Robinson 1978). It has been recommended that specimens for penicillin assay should be stored at −70°C and assayed within 90 days (Kaplan et al 1989). We conducted a trial to document the rate of degeneration of penicillin in specimens stored at −20°C for up to 84 days as part of a study on rheumatic fever prophylaxis in South Africa (Daniels et al 1994).

Serum and urine specimens were collected from eight patients receiving benzathine penicillin (Penilente 1·8 g; Novo Industries) as secondary prophylaxis for rheumatic fever. Specimens were collected 28 days after intramuscular administration of 1·2 m units benzathine penicillin. Six samples (0·5 mL) of serum and of urine were prepared from each of the eight patients and were then frozen to −20°C. Specimens were stored for up to 84 days. A single sample of serum and of urine from each patient was thawed at 2-weekly intervals and the penicillin assay performed. The microbiological agar diffusion method involves incubating duplicate samples of specimens placed in wells in the agar plate which is impregnated with a penicillin-sensitive organism (Sutherland & Rolinson 1978). Duplicate samples of 5–6 standard solutions of penicillin are also placed on the plate. Penicillin concentrations are derived by measurement of the zones of growth inhibition of the organism around each well. The lower limit of sensitivity of the assay was 0·015 $\mu g\ mL^{-1}$. Penicillin concentrations lower than this were, therefore, recorded as undetectable.

Because of the large number of undetectable serum penicillin concentrations, the relationship between penicillin concentrations and duration of storage was tested for the urine samples only. Penicillin was detected in the urine of all eight patients. The mean coefficient of variation for the urine penicillin concentrations was 16%. There was no significant difference in the mean urine penicillin concentrations between the six assays ($P = 0.99$) (Table 1).

Table 1. Results of serial determinations of urine penicillin concentrations ($\mu g\ mL^{-1}$) for eight patients.

Patient	Number of days between collection of specimens and assay						CV* (%)
	14	28	42	56	70	84	
1	9·50	9·00	7·50	7·50	11·50	10·00	16·8
2	5·25	3·20	5·25	3·25	4·00	4·50	21·7
3	20·00	23·00	18·00	19·50	17·50	25·00	14·3
4	1·23	1·40	1·55	1·75	1·12	0·82	25·1
5	2·62	3·50	3·00	3·00	2·75	3·12	10·2
6	0·50	0·55	0·52	0·47	0·40	0·45	11·1
7	1·85	2·10	2·50	1·60	1·45	1·70	20·4
8	13·50	17·00	13·50	14·50	15·50	18·00	12·1
Mean %	6·81	7·45	6·48	6·45	6·78	7·95	

We therefore conclude that urine specimens for penicillin assay can be stored at −20°C for up to 84 days if the preparation administered is benzathine penicillin. This informa-

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tion may be especially important for investigators in developing countries where rheumatic fever remains a public health problem and where facilities for storage of specimens at −70°C may not be readily available.

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References

Daniels, E. D., Mohanlal, D., Pettifor, J. M. (1994) Rheumatic fever prophylaxis in South Africa-is bicillin 1·2 mu 4-weekly appropriate? S. Afr. Med. J. 84: 477–481.

Kaplan, E. L., Berrios, X., Speth, J., Siefferman, T., Guzman, B., Quesny, F. (1989) Pharmacokinetics of benzathine penicillin G: serum levels during the 28 days after intramuscular injection of 1200000 units. J. Pediatr. 115: 146–150.

Sutherland, R., Rolinson, G. N. (1978) Methods of antibiotic assay. In: Reeves, D. S., Philips, I., Williams, J. D., Wise, R. (eds) Laboratory Methods in Antimicrobial Therapy, Churchill Livingstone, pp 171–178

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Book Review

Trends and Future Perspectives in Peptide and Protein Drug Delivery

(Drug Targeting and Delivery Volume 4)

Edited by Vincent H. L. Lee, Mitsuru Hashida and Yutaka Mizushima

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To be the editor or co-editor of a volume of conference proceedings is not an easy task. The usefulness of such a collection of papers is dependent on rapid publication such that the volume represents state-of-the-art technology. One practical way of reducing printing and publication time is to prepare the book from camera ready copy (CRC). It is usual for the authors to be given a set of instructions, which contain guidelines on the typeface, font size, spacing etc., in order to generate some uniformity. A scan of this volume, however, suggests that many of the authors were either not in possession of manuscript preparation guidelines or chose to ignore them. The first two chapters illustrate the case in point. Chapter 1, the sole occupant of section I, presents an overview of peptide and protein drug delivery written by Lee, one of the editors. Although the contents of this chapter do provide a reasonable introduction to the remainder of the volume, the typeface and layout of the text make it an extremely difficult read. If this was not enough, the chapter is riddled with grammatical and typographical errors, including the classical misspelling of the word "accommodate". This reviewer is also somewhat puzzled by some of the author's statements such as "...oral route has to make room for mucosal routes..." and "...all the mucosal routes suffer from a low surface area for absorption...". Although my physiology classes were taken some twenty-five years ago, I do recall that the gastrointestinal surface was described as mucosal and that the lining of the duodenum, jejunum and ileum were specially adapted to provide a large surface area for absorption!

In complete contrast to chapter 1, chapter 2 (by Peppas) illustrates just how good CRC can be. This chapter is well set out and quite readable, although I have to admit to consulting my dictionary to look up the meaning of the word "reptation". As a general up-to-date introduction to protein diffusion in hydrogels it is invaluable. The remaining two chapters of Section II cover the use of poly(ortho esters) for the delivery of peptides and proteins and new delivery systems for recombinant proteins. Chapter 3, written by industrial scientists from Genentech Inc., illustrates the problems that may be encountered in developing the right delivery system for specific proteins and gives much food for thought—it should be read by every pharmaceutical scientist about to enter the protein drug arena.

Section III of this volume is entitled "Transport Enhancement of Peptide and Protein Drugs Across Selected Absorptive Barriers". There are five chapters which cover transport enhancement across the gastrointestinal tract, skin and the blood–brain barrier. Iontophoretic delivery, the most promising mechanism for enhancing peptide permeation through skin, is described in two chapters; the contribution by Sage et al providing true insight into practical issues. The remaining three sections cover targeting, the methods used to improve the pharmacokinetic and pharmacodynamic properties of peptide and protein drugs, and delivery of oligonucleotides and genes.

Provided the frustration generated by continually changing typefaces can be overcome, the reader will find that this volume contains a collection of useful and interesting chapters, provided by leaders in their specialist fields. The balance of contributions from academia and industry ensures that both the basic science and the practical developmental issues are covered in depth. There should be a copy in the libraries of those institutions working in the area of peptide and protein drugs.

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