

**SEMI-MICRO DETERMINATION OF WATER CONTENT FOR THE BULK
SAMPLES OF OXYTETRACYCLINE, TETRACYCLINE AND
CHLORTETRACYCLINE**

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ABSTRACT

The semi-micro analysis of the bulk commercial samples of Oxytetracyclin, Tetracycline and Chlortetracycline for their water content using Karl Fischer reagent is described. All the samples analysed comply with the limits laid down in European Pharmacopoeia or United States Pharmacopoeia.

Introduction

The overaged tetracycline preparations in general are degrading to 4-epianhydrotetracyclines (EATCs) which shows a definite toxicity to renal tubular function (Fanconi Syndrome), (Benitz and Diermeier, 1964); Gross, 1963 and Frimpter *et al*, 1963). The United States Pharmacopoeia (U.S.P.) limits EATC in preparations to 3% and in bulk samples upto 2% Rockville (1985).

However the excessive water content influences the formation of acid catalysed decomposition products. The elevated temperature and excessive moisture content are said to be responsible for the formation of EATCS. The work on the stability of oxytetracycline (OTC) and tetracycline (TC) has already been published describing the vital role of moisture content in the formation of EATC, (Khan *et al*, 1987 and Dihuidi *et al*, 1982). It can therefore be concluded that increase of water content facilitates the formation of decomposition products among the tetracyclines group of antibiotics.

Experiments:

Solvents and reagents

Karl Fischer reagent from Merck Darmstd, F.R.G. Methanol was the HPLC grade from Ruthburns, U.K.

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Apparatus

Metrohm 633 Karl-Fischer Automat (Metrohm, Herisau, Switzerland) equipped with a 645 Multi-Dosimat and a multi-Burette E 485. Specially glass fabricated sample weighing bottles with covers were purchased from the local market.

Results and discussions

Semi-micro determination of water was carried out following the instructions of European Pharmacopoeia Vol. 3.5.6, 1980. The weighing bottles were washed with water, rinsed with ethanol and dried in an oven at 120°C for 2 hours. Then these bottles were cooled down in a dessicator over diphosphorus pentoxide. The sample was weighed in a weighing bottle and closed immediately with the cover to avoid penetration of moisture into the sample weighed. The sample was then transferred into the automatic titration flask through a special chamber and carried out the titration.

Table 1**BULK SAMPLES OF OXYTETRACYCLINE BASE**

Value in percent (m/m) expressed as the base; NM = not mentioned.

Sample Origin	Sample number	Sample age in months	Water content (Karl Fischer)		
			Number of analyses	Mean %	RSD %
Manufacturer					
A	23	NM	3	7.4	0.7
	24	NM	3	6.8	0.5
F	63	NM	3	6.5	0.3
	64	NM	3	6.2	<0.1
G	68	NM	3	7.4	0.2
H	75	NM	3	7.4	1.0
	77	NH	3	7.9	0.4
Whole-Salers	66	NM	3	7.7	0.4
	71	13	3	7.9	0.3

Table 1 shows the results of commercial OTC hydrated samples. All the samples give values between 6A and 8.0 percent. The theoretical water content for OTC dihydrate is 7.8%. The European Pharmacopoeia (Ph. Eur.) 1983 limits the water content to 4.0-8.0% and the United States Pharmacopoeia, 1985 to 6.0-9.0%. The results for the bulk samples of OTC hydrochloride (OTC. HCl) salt is presented in Table 2.

Table 2**BULK SAMPLES OF OXYTETRACYCLINE HYDROCHLORIDE**

**VALUE IN PERCENT (m/m) expressed as the hydrochloride;
NM = not mentioned.**

Sample origin	Sample number	Sample age in months	Water content (Karl Fischer)		
			Number of analyses	Mean %	RSD %
Manufacturer					
A	20	NM	4	1.6	3.2
G	69	NM	5	0.9	3.0
	70	NM	4	0.7	2.7
H	74	NH	5	0.8	2.6
I	78	NM	4	6.6	1.9
K	72	9	8	0.8	4.2
L	73	9	3	0.8	2.5
N	31	NM	4	7.7	0.7
Whole Salers					
	1	60	4	6.5	0.8
	4	35	6	0.8	2.6
	6	38	4	7.5	3.7
	9	38	7	3.9	1.5
	11	38	4	3.8	3.4
	14	NM	3	7.2	0.8
	25	NM	3	7.3	0.8
	27	NM	3	0.8	0.7

The Ph. Eur. and U.S.P. limits the water content to 2.0%. The limit is exceeded in many samples. This may be due to the manufacturing process or misconservation. Some of the OTC. HCl bulk samples were also found to be hygroscopic. It has been described that OTC. HCl samples can appear in three polymorphic modifications. Two of the three

described polymorphs are hygroscopic (Burger, 1986). This might be the reason for the variation in hygroscopicity and higher limits of the water content in the OTC. HCl samples. Here it is beyond the scope of this study to investigate the relationship between the moisture content of the samples and the crystalline structures. The relative standard deviations (R.S.D.) in shows the good performance of the analyses.

Table 3 shows the results for bulk samples of Tetracycline base (TC) represented in the same way as for OTC. The Ph. Eur. and U.S.P. limit the water content to 13%. Water is found to be within limits for all the samples. For the bulk samples of TC hydrochloride salt (TC. HCl), the results are given in Table 4. The water content was found to be within Ph. Eur. and U.S.P. limits i.e. 2.0%.

Table 3

BULK SAMPLE OF TETRACYCLINE BASE

Values in percent (m/m) expressed in terms of the base; Sample age not mentioned on the label.

Manufacturer	Sample number	Water Content (Karl Fischer)		
		Number of analyses	Mean %	RSD %
A	27	3	10.3	0.3
B	57	3	6.1	1.4
	58	3	3.2	1.8
	59	3	5.2	1.9
C	60	3	6.2	1.9
	61	3	8.2	0.4
	62	3	3.7	4.5
	63	3	7.3	0.4
D	64	3	8.1	0.8

Table 5 shows the results obtained for the bulk samples of chlortetracycline hydrochloride salt (CTC. HCl). The official texts prescribe the loss on drying instead of Karl Fischer titrations. But owing to the limited amount of samples available, Karl Fischer titrations were carried out because loss on drying requires 1.00g of the sample for each analysis while for the determination of water by Karl Fischer titration the weight required is only 0.10g.

Table 4

BULK SAMPLES OF TETRACYCLINE HYDROCHLORIDE

Values in percent (m/m) expressed in terms of the hydrochloride
 NM = not mentioned.

Sample Origin	Sample number	Sample age in months.	Water content (Karl Fischer)		
			Number of analyses	Mean %	RSD %
Manufacturer B	1	NM	3	1.2	0.9
Whole Salers					
1	5	28	3	0.9	1.2
	10	41	3	1.4	2.6
	12	49	3	1.1	2.1
	19	52	3	1.6	4.1
	20	32	5	1.3	6.3
	23	20	3	1.1	0.5
2	2	30	3	1.4	0.8
3	3	16	3	1.0	2.6
	15	31	3	1.0	0.6
	16	40	2	1.8	1.6
4	17	44	4	1.0	7.2
	48	22	3	0.8	2.5
	6	38	3	1.2	4.8
5	4	NM	3	0.9	1.8
6	18	NM	3	1.4	2.9
7	21	NM	3	0.9	2.7
8	50	9	3	0.9	1.7
	51	6	4	0.6	4.2
9	49	28	3	0.8	3.3

Table 5

BULK SAMPLES OF CHLORTETRACYCLINE HYDROCHLORIDE:

Values in percent (m/m) expressed in terms of the hydrochloride; Sample age not mentioned on the label.

Sample Origin	Sample number	Water content (Karl Fischer)		
		Number of analyses	Mean %	RSD %
Manufacturer				
A	2	3	0.7	1.5
	3	3	0.4	2.7
	4	3	0.6	5.1
	5	3	0.3	9.1
	14	3	1.8	1.6
	15	3	1.1	4.3
B	8	3	0.5	4.2
Whole Salers				
1	6	3	0.4	1.3
	7	3	0.8	8.5
	11	3	0.5	5.0
2	1	3	0.5	6.0
	9	3	0.6	1.0
	12	3	0.8	0.7
3	10	3	0.5	3.7
4	13	3	0.8	3.4

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