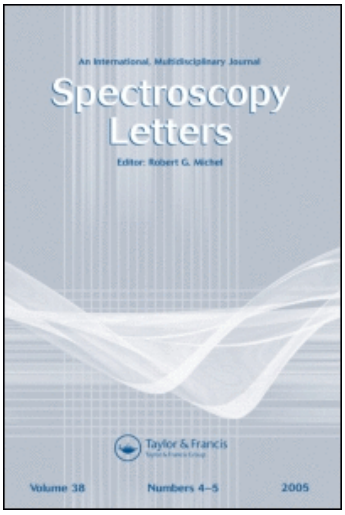

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Spectrophotometric Determination of Niclosamide Using P-Benzoquinone

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SPECTROPHOTOMETRIC DETERMINATION OF NICLOSAMIDE USING P-BENZOQUINONE

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Key Words: Niclosamide Determination; P-benzoquinone; spectrophotometry

ABSTRACT

A simple spectrophotometric method for the determination of niclosamide in pure and dosage forms has been developed. The proposed method is based on the reduction of the nitro group in niclosamide to the amino group by heating in a water bath a mixture of niclosamide solution in 95% ethanol, zinc powder and dilute hydrochloric acid for 15 minutes. The cold and clear filtrate reacts with p-benzoquinone, where a compound of pink colour is obtained which absorbs maximally at 506 nm.

The stoichiometry of the proposed method and the possible pathway of the reaction is presented. The method determines from 12.5-125 μg/ml of niclosamide with mean percentage recovery of 100.07 ± 0.707.

The suggested method was applied to Niclosan and Yomesan tablets and its validity was ascertained by standard addition technique.

INTRODUCTION

Niclosamide is used for the treatment of tapeworm infestations. Various methods including spectrophotometry[1-10], gas chromatography[11,12], HPLC[13,14] and polarography[15] have been used for the determination of niclosamide.

Reaction of primary and secondary amines with P-benzoquinone has been used as the basis for spectrophotometric determination of considerable number of amine containing medicinal compounds[16-18]. Niclosamide on reduction gives a product containing a primary amino group which can reacts with P-benzoquinone to form a pink complex. In this work the reduction of niclosamide as well as the reaction of its reduced product with P-benzoquinone has been studied to declare the optimum reaction conditions such as temperature, time and reagent concentration for each reaction. It was found that the reaction product with p-benzoquinone can be measured spectrophotometrically. The proposed spectrophotometric method was adopted for determination of niclosamide in pure samples as well as in its dosage forms.

EXPERIMENTAL

Samples, Reagents and Apparatus

Samples

1- Pure sample of niclosamide; provided by MISR Co. For PHARM. IND. S.A.A., Cairo A.R.E. It was found to Contain 99.43 ± 0.566 according to E.P. (1984)[19].

Reduced Niclosamide standard solution; was prepared in concentration 0.25 mg/ml by heating 62.5 mg of niclosamide with 100 ml hot 95% ethanol, 100 mg zinc powder and 5 ml dilute hydrochloric acid in a boiling water bath for 15 minutes, then cool filter and wash the residue with 95% ethanol. Collect the filtrate and the

washing into 250 ml measuring flask and complete to the mark with 95% ethanol.

2- Commercial samples

a- Niclosan tablets, a product of MISR Co. for PHARM IND. S.A.A., Cairo. A.R.E. Batch No 130034 and 152066, each tablet is claimed to contain 500 mg niclosamide.

b- Yomesan tablets, a product of BAYER Turk Kimya San. Ltd. Sti. Batch No 503191 and 508197, each tablet is claimed to contain 500 mg niclosamide.

Reagents

All chemicals were of analytical grade.

1- P-benzoquinone solution; was prepared in concentration of 8 mg/ml in 95% ethanol.

2- Ethanol 95% v/v, distilled over potassium hydroxide before use.

3- Zinc powder; 99%.

Apparatus

Beckman Du-7 spectrophotometer, calibrated before use and connected to printing recorder.

Procedures:

A. For bulk powder

Into a series of 20 ml test tubes, transfer aliquots of the reduced niclosamide standard solution equivalent to 125-1250 μg, add one ml p-benzoquinone reagent, complete the volume to 6 ml with 95% ethanol. Mix well and heat in a boiling water bath for 15 minutes. Leave to cool, transfer into 10 ml measuring flasks and complete to the mark with 95% ethanol. Measure the absorbance at 506 nm against a reagent blank.

B- Determination of niclosamide in tablets

Accurately weigh and finely powder 10 tablets. Weigh accurately a quantity of powdered tablets equivalent to 62.5 mg of niclosamide. The reduction of niclosamide was carried out as mentioned before under preparation of reduced niclosamide standard solution. Take different aliquots of the final solution equivalent to 125-1250 μg of the reduced niclosamide and proceed as stated under the procedure for bulk powder. The standard addition technique was applied to check the validity of the suggested spectrophotometric determination of niclosamide in the tablets.

RESULTS AND DISCUSSION

Zinc powder in dilute hydrochloric acid is used as reducing agent to reduce the nitro group in niclosamide to the amino group, heating the reactants in a boiling water bath for 15 minutes initiates the reduction process[19], more over, the addition of 95% ethanol assists in the solubilization of niclosamide and its reduced product[20]. This preformed amino group can reacts with p-benzoquinone, to get the highest sensitivity of the reaction product is attained by heating the reactants on a boiling water bath at 100°C for 15 minutes. The optimum concentration of p-benzoquinone used is found to be 1 ml of 8 mg/ml solution.

The product of the reaction between the reduced niclosmide and p-benzoquinone has a pink colour which absorbs maximally at 506 nm (Fig 1).

The study of the stoichiometry of the reaction by applying the mole ratio method, showed, that the ratio of reactants is 1:1.

Isolation of Reaction product

The reduction process of niclosamide was carried out as mentioned before using 50 mg of niclosamide. The reaction mixture

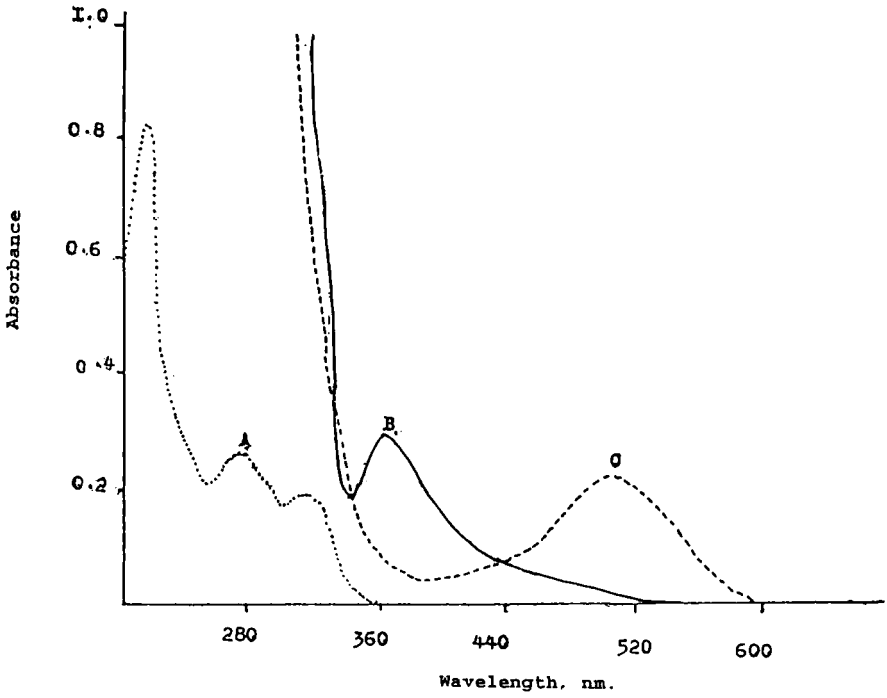

Fig. 1: Absorption Spectra of:
A- The Reduction Product of Niclosamide (0.1mg/ml)
B- P-Benzoquinone (0.08 mg/ml)
C- Reaction Product of the Reduced Niclosamide (0.025 mg/ml) with P-Benzoquinone.

was heated in a boiling water bath for 15 minutes. After cooling the solution was filtered to remove excess zinc powder. The reduced niclosamide was precipitated by addition of excess distilled water, heated in a boiling water bath to coagulate the precipitate and centrifuged. The residue was recrystallized from hot 95% ethanol the product obtained was identified by I.R.

Another amount equal to 50 mg of niclosamide was reduced as stated in the previous paragraph. The clear filtrate containing the reduced niclosamide was reacted with about 320 mg of p-benzoquinone in 95% ethanol. The reaction mixture was heated in a boiling water

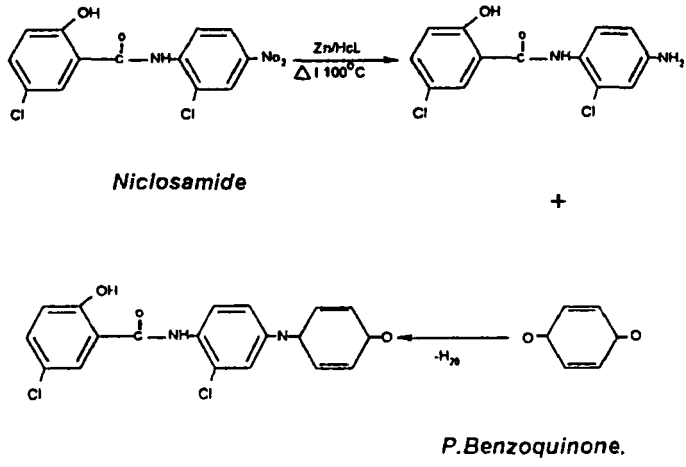

Fig. 2: The Suggested Mechanism of The Reaction Between Niclosamide and P.Benzoquinone.

bath for 15 minutes. After cooling the reaction product was precipitated by addition of excess distilled water and the precipitate was treated as mentioned before to identify by I.R.

The IR spectrum of the isolated reduced niclosamide shows two bands at 3574.7 and 3487.4 cm^{-1} due to N-H stretching for primary aromatic amines, while these two bands are disappeared in the IR spectrum of the isolated reaction product between reduced niclosamide and p-benzoquinone. This proves that the reduced niclosamide reacts through its amino group with the carbonyl group of p-benzoquinone to form the pink coloured product. From the study of the stoichiometry of the reaction and IR spectrum the mechanism of reaction can be represented in Fig 2.

For the proposed method Beer's law is obeyed in the concentration range of 12.5-125 μg/ml. The regression equation is $A = 8.85 \times 10^{-3} C - 7.00 \times 10^{-4}$ where A is absorbance and C is concentration in μg/ml.

Table 1: Determination of pure sample of niclosamide using the proposed method.

Exp.No	Taken mg/ml	Found mg/ml	Recovery %
1	12.5	12.35	98.80
2	25.0	25.21	100.84
3	50.0	50.30	100.60
4	75.0	75.03	100.04
5	100.0	100.13	100.13
6	125.0	124.98	99.98
Mean ± S.D	100.07 ± 0.707		

Table 2: Statistical comparison between the proposed Method and the E.P. method [19].

Preparation		Proposed methed	Pharmacopoeial methed
Pure sample of niclosamide	Mean ± S.D.	100.07 ± 0.707	99.43 ± 0.566
	n	6	5
	variance	0.500	0.320
	t	1.631 (2.262)	
	f	1.563 (6.26)	
Niclosan tabletes B.N.130034	Mean ± S.D.	99.98 ± 0.465	100.04 ± 0.716
	n	5	5
	variance	0.216	0.513
	t	0.157 (2.306)	
	f	2.375 (6.39)	
Niclosan tabletes B.N.152066	Mean ± S.D.	100.26 ± 0.451	99.9 ± 0.782
	n	5	5
	variance	0.203	0.612
	t	0.867 (2.306)	
	f	3.015 (6.39)	
Yomesan tablets B.N.503191	Mean ± S.D.	99.56 ± 0.457	99.42 ± 0.561
	n	5	5
	variance	0.209	0.315
	t	0.432 (2.306)	
	f	1.507 (6.39)	
Yomesan tablets B.N.508197	Mean ± S.D.	99.14 ± 0.722	99.35 ± 0.581
	n	5	5
	variance	0.521	0.338
	t	0.507 (2.306)	
	f	1.541 (6.39)	

The values in parentheses are the tabulated t and f values.

Table 3: Determination of niclosamide in pharmaceutical preparations using the proposed method.

Niclosan Tablets *		Standard Addition			Yemesan Tablets **		Standard Addition		
Claimed mg/tablets	found*** %	added µg/ml	found µg/ml	recovery %	Claimed mg/tablets	found*** %	added µg/ml	found µg/ml	recovery %
500	99.98 ± 0.465	25	25.18	100.72	500	99.56 ± 0.457	25	24.99	99.96
		50	49.90	99.80			50	49.85	99.70
		75	75.34	100.45			75	74.81	99.75
		100	99.70	99.70			100	100.05	100.05
				100.17 ± 0.496					99.87 ± 0.167

During application of the proposed method for determination of pure samples of niclosamide, the mean percentage recovery was found to be 100·07 ± 0.707 (Table 1).

The results obtained by applying the proposed method to determine niclosamide in pure samples and in Niclosan and Yomesan tablets were compared statistically to those obtained by the pharmacopoeial method. Table 2 shows that the "f" and "t" values are less than the corresponding theoretical values indicating that there is no significant difference between the two methods with respect to both precision and accuracy.

The validity of the proposed method was checked by applying standard addition technique. Table 3. shows that the percentage recovery of the added niclosamide has almost the same accuracy as those previous obtained with pure samples of niclosamide.

Finally the proposed method has the advantage in being so simple, inexpensive, sensitive and applicable over a convenient range of niclosamide concentration.

REFERENCES

1- N.M. Sanghavi, V.H. Sathe and M.M. Padki; Indian Drugs, 20 (8) 341-342 (1983) (Anal Abstr). 45 (4) 4E3 (1983).

2- N. Bergisadi and D. Sarigul; Acta Pharm. Turc., 28 (2) 51-54 (1986) (Anal. Abstr. 48 (12) 12E 63 (1986).

3- G. Yang and T. Shi; Yaowu Fenxi Zazhi, 7 (6) 374 (1987) (Anal. Abstr. 50 (7) 7E 92 (1988).

4- C.S.P. Sastry, M. Aruna, A. Rao and M.Rama; Indian Drugs 25 (8) 348-349 (1988) (Anal. Abstr. 50 (10) 10E 84 (1988).

5- C.S.P. Sastry ,M, Aruna, A. Rao and M. Rama; Talanta, 35 (1) 23-26 (1988).

6- S.S. Zarapkar, S.R. Mehra and R.V. Rele; Indian Drugs 26 (7) 360-362 (1989) (Anal. Abstr. 52 (1) 1E 41 (1990).

7- B.V. Lopatin, N.B. Lopatina and A.F. Bekhli; Farmatsiya (Mascow), 38 (3) 75--76 (1989) (Anal. Abstr. 52 (3) 3E 53 (1990).

8- C.S.P. Sastry M. Aruna, A. Rao, M. Rama and A.S.R.P. Tripireni; Chem. Anal. Warsaw, 36 (1) 153-158 (1991).

9- L.H. Emara; J. AOAC. Int., 76 (4) 847-850 (1993).

10- F. Onur and N. Tekin; Anal. Lett. 27 (12) 2291-2301 (1994).

11- T. Daldrup, F. Susanto and p. Mickalke; Fresenius Z. Anal. Chem, 308 (5)-413 (1981).

12- N.Nose, S. Kobayashi, A. Tanaka, A. Hirose and A. Watanabe; J. Chromatogr., 130, 410 (1977).

13- T. Daldrup, P. Mickalke and W. Boehme; J. Chromatogr. Newsl, 10 (1) 1 (1982).

14- O.M. Solodka and G.A.Donets; Farm. Zh. (kiev), Nov-Dec (6) 58-59 (1990) (Anal-Abst. 54 (3) 3E 132 (1992).

15- A.K. Mishra and K.D. Gode; Indian Drugs, 22 (6) 317-320 (1985) (Anal. Abstr. 48 (2) 2E 58 (1986).

16- M.F. Loucks and L. Nauer; J. Assoc. Off. Anal. Chem. 50, 268 (1968).

17- G.A. Benson and W. Spillane; J. Anal. Chem., 48, 2149 (1976).

18- A.M. Wahbi, M.A. Abounassif and E.A. Gadkariem, Talanta, 33, 179 (1986).

19- The English Text of the Egyptian Pharmacopeia, 3rd Edition volum 1, 578 (1984).

20- British Pharmacopoeia, Volum II, 445 (1993).

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