



QUANTITATIVE ANALYSIS OF DIFFERENT BRANDS OF DEXAMETHASONE TABLET MARKETED IN MAIDUGURI METROPOLITAN COUNCIL

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ABSTRACT

This experiment involves quantitative analysis of dexamethasone tablet marketed in Maiduguri Metropolitan Council using UV-Spectrophotometry and HPLC method of analysis in which the samples were dissolved in methanol and their absorbance were determined and compared to that of the standard. Percentage content for each of the samples was determined so as to note if it was within the acceptable range of 90-110% in the British Pharmacopoeia (BP), for those that passed the test or, if it was below or above the range for samples that are substandard or highly concentrated. The amount of dexamethasone tablet in each sample was determined (actual content) and compared with that of the stated amount (0.5mg). It was observed that out of the eight samples (AARON'S, KRIS, NKOYO, DEXAREL, LOKEL'S, ANISUN, CHAZMAX and DEXA) three of the samples (NKOYO, LOKEL'S and DEXA) met the BP requirement for actual drug content range of 90-110% with percentage content of 98.1, 99.2 and 95.8% respectively, while the other five samples failed the test with values below and above the acceptable range.

Keyword: Dexamethasone, Tablet, HPLC and UV-Spectrophotometer

INTRODUCTION

There is a global concern on the rapid increase of substandard and adulterated pharmaceutical products in many countries of the world including Nigeria¹. The emergence of substandard pharmaceutical products (drugs) in Nigeria can be attributed to many factors which may include; Inadequate funding of hospital Pharmacies and the "out of stock syndrome", Involvement of unqualified persons in the procurement and distribution of drugs, inadequate storage facilities, transportation and distribution amongst others².

Nigeria as a country imports its drugs from countries like the US, EU and China to meet its healthcare needs. The world health organization (WHO) has recommended that countries who engage in the importation of drugs should protect their countries from the danger of substandard drugs by ensuring that drugs imported are properly sampled and analysed³. It is the responsibility of the government to protect its citizens from the clutches of unscrupulous members of the society. The Nigerian government has designed various ways to do this, and it is expected to equip the regulatory agencies with materials and manpower to effectively perform their duties⁴.

GLUCOCORTICOIDS

Glucocorticoids (GC) of which dexamethasone are a synthetic derivative are a class of steroid hormones that bind to the glucocorticoid receptor (GR), which is present in almost every vertebrate animal cell. The name glucocorticoid (glucose + cortex + steroid) derives from their role in the regulation of the metabolism of glucose, their synthesis in the adrenal cortex, and their steroidal structure¹.

Glucocorticoids are part of the feedback mechanism in the immune system that turns immune activity (inflammation) down. They are used in medicine to treat diseases that are caused by an overactive immune system, such as allergies, asthma, autoimmune diseases and sepsis. Glucocorticoids have many diverse effects, including potentially harmful side effects⁶. They interfere with some of the abnormal mechanisms in cancer cells, so they are used in high doses to treat cancer.

Various synthetic glucocorticoids are available; Betamethasone, Dexamethasone, these are used either as replacement therapy in glucocorticoid deficiency or to suppress the immune system.

THEORY OF ANALYTICAL METHODS

UV- visible Spectrophotometry

The fundamental application of ultraviolet and visible spectrophotometry is in quantitative analysis. It can be used for assay of single, mixture and bulk drugs, dissolution tests for tablets and capsules, limit test for impurities (colorimetric methods). Also, it can be used for measurements such as pKa or velocity constants in enzymatic reactions⁷. A UV spectrophotometer may be used as a detector for HPLC. The presence of an analyte gives response assumed to be proportional to the concentration. For accurate results, the instrument's response to the analyte in the unknown should be compared with the response to a standard.

High performance liquid chromatography (HPLC)

The ability to separate and analyse complex samples is integral to the pharmaceutical and medical science. Classic column chromatography has evolved over the years with chromatographic innovations introduced at roughly decade intervals. These techniques offered major improvements in speed, detection, quantification, resolving power, convenience and applicability to new sample types. The most notable of these modifications was high performance liquid chromatography (HPLC)⁸.

High performance liquid chromatography (sometimes referred to as high pressure liquid chromatography) is a chromatographic technique that can separate a mixture of compounds and is used in biochemistry and analytical chemistry to identify, purify and quantify the individual components of a mixture⁹.

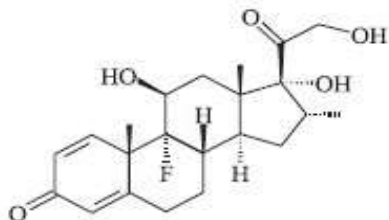
The predominant method in the pharmaceutical analysis of steroids (e.g. dexamethasone) is the reversed phase HPLC with UV detection, but the technique is time consuming, costly and requires expertise¹⁰.

In May 2010, Joseph Emil Amegadzie carried out a quantitative analysis on dexamethasone tablets using the HPLC method of analysis. In this analysis, the percentage purity of dexamethasone when it was analysed was 99.40%. The British pharmacopoeia states that, its percentage purity ought to be between 90.0% and 110.0%¹¹. Based on his result, the sample analysed contained the amount required as stated by the British Pharmacopoeia and that the HPLC method was the appropriate technique to employ for the analysis.

In September 2009, Rossana et al. gave a report on the validation of an analytical UV Spectrophotometric method to assay dexamethasone tablets. In their report, three different samples of dexamethasone tablets were used and the percentage drug content obtained were; 99.34%, 99.88%, and 98.49%. The results obtained were in accordance with the official specifications of between 90.0% and 110.0%¹¹.

DEXAMETHASONE AS A GLUCOCORTICOID.

Dexamethasone, 9 α -fluoro-16 α -methyl-11 β , 17 α , 21-trihydroxy-1,4-pregnadiene-3,20-dione, is a synthetic derivative of the glucocorticoid hydrocortisone that has a long history of use in human, it is a potent synthetic member of the glucocorticoid class of steroid drugs. It acts as an anti-inflammatory and immunosuppressant. It is 20 to 30 times more potent than the naturally occurring hormone cortisol and 4 to 5 times more potent than prednisone¹². Dexamethasone binds more powerfully to the glucocorticoid receptor than cortisol does. Dexamethasone is based on the cortisol structure but differs in three positions (extra double bond in the A-ring between carbons 1 and 2 and addition of a 9- α -fluoro group and a 16- α -methyl substituent)¹³.



Chemical structure of dexamethasone B

Chemical data¹⁴

Formula $C_{22}H_{29}FO_5$

Mol. Mass 392.461 g/mol

MATERIALS AND METHOD

REAGENTS

Methanol

Distilled water

SAMPLE COLLECTION

Eight (8) different brands of dexamethasone tablets were collected from different locations within Maiduguri metropolis. Their various batch number, NAFDAC registration number, expiry date, manufacturing date and labelled strength were documented¹¹.

METHODS USED FOR THE ANALYSIS OF DEXAMETHASONE TABLET

UV spectrophotometry

High performance liquid chromatography (HPLC)¹⁶

Procedure;

The weight of ten (10) tablets from each sample brand was taken singly and then the average weight determined.

Each of the samples was crushed using the mortar and pestle, the required amount of the sample needed was weighed.

Methanol (MEOH) was prepared and poured into a 500ml volumetric flask

50ml volumetric flasks were used to put the different samples and each flask was labelled appropriately.

Small amount of the prepared methanol was then added and made up to the 50ml mark in each flask.

The solutions were sonicated for 15min using a sonicator and then filtered to get a clear solution.

2.5ml of the filtrate was withdrawn from each volumetric flask using a 5ml pipette, poured into a new 50ml conical flask and made up to the 50ml mark.

Small amount was taken from each flask and put into a cuvette.

The same procedure was then repeated for the standard dexamethasone.

The absorbance was taken using the UV- spectrophotometer at a wave length of 238nm. Also the samples were placed into the HPLC machine for analysis.

The percentage content and milligram content of each of the samples was then determined and compared with the standard¹⁷.

RESULTS

TABLE 1: SHOWING INFORMATION ABOUT THE VARIOUS SAMPLES

BRAND	BATCH NO:	MANUFACTURING DATE	EXPIRATION DATE	NAFDAC REG. NO:	LABELED STRENGTH
AARON'S	DE-06	MAY 2010	APRIL 2013	KD-302	0.5
KRIS	E-105	MAY 2011	APRIL 2015	A4-0685	0.5
NKOYO	T-10405	APRIL 2010	MARCH 2013	04-8665	0.5
DEXAREL	DES-107	FEB. 2011	JAN. 2015	04-4641	0.5
LOKELS	C-111	MARCH 2011	FEB. 2015	04-7854	0.5
ANISUN	TMF-11	JUNE 2011	MAY 2014	04-8422	0.5
CHAZMAX	DX-001	SEPT. 2010	AUG. 2014	A4-2197	0.5
DEXA	MXT-15	MARCH 2010	FEB. 2013	KD-1912A	0.5

TABLE 2: SHOWING THE AVERAGE WEIGHTS OF THE VARIOUS SAMPLES OF DEXAMETHASONE TABLETS IN Mg.

BRAND	MINIMUM WEIGHT	MAXIMUM WEIGHT	AVERAGE WEIGHT
AARON'S	70	80	75
KRIS	70	70	70
NKOYO	60	80	70
DEXAREL	70	80	74
LOKEL'S	70	70	70
ANISUN	60	80	69
CHAZMAX	80	100	86
DEXA	60	80	76

Calculation of percentage content of the samples using UV-spectrophotometry at a wavelength of 238nm.

$$\% \text{ Content} = \frac{\text{Absorbance of sample}}{\text{Absorbance of standard}} \times 100\%$$

STANDARD (absorbance = 1.425.)

$$\% \text{ Content} = \frac{1.425}{1.425} \times 100\% = 100\%$$

AARON'S (Absorbance = 1.656)

$$\% \text{ Content} = \frac{1.656}{1.425} \times 100\% = 116.2\%$$

KRIS (Absorbance = 1.135)

$$\% \text{ Content} = \frac{1.135}{1.425} \times 100\% = 79.6\%$$

NKOYO (Absorbance = 1.398)

$$\% \text{ Content} = \frac{1.398}{1.425} \times 100\% = 98.1\%$$

DEXAREL (Absorbance = 1.221)

$$\% \text{ Content} = \frac{1.221}{1.425} \times 100\% = 85.7\%$$

LOKEL'S (Absorbance = 1.414)

$$\% \text{ Content} = \frac{1.414}{1.425} \times 100\% = 99.2\%$$

ANISUN (Absorbance = 1.134)

$$\% \text{ Content} = \frac{1.134}{1.425} \times 100\% = 79.6\%$$

CHAZMAX (Absorbance = 1.232)

$$\% \text{ Content} = \frac{1.135}{1.425} \times 100\% = 86.5\%$$

DEXA (Absorbance = 1.365)

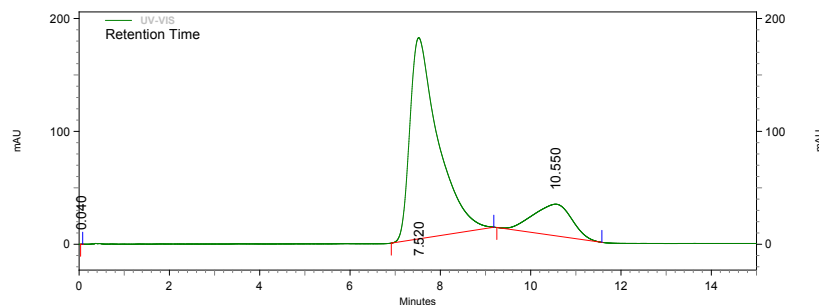
$$\% \text{ Content} = \frac{1.365}{1.425} \times 100\% = 95.8\%$$

TABLE 3: SHOWING THE PERCENTAGE CONTENT OF THE SAMPLES USING UV-SPECTROPHOTOMETRY AT A WAVE LENGTH OF 238nm

BRAND	ABSORBANCE	% CONTENT
STANDARD	1.425	100
AARON'S	1.656	116.2
KRIS	1.135	79.6
NKOYO	1.316	98.1
DEXAREL	1.221	85.7
LOKEL'S	1.414	99.2
ANISUN	1.134	79.6
CHAZMAX	1.232	86.5
DEXA	1.365	95.8

Analyst: LAB MANAGER

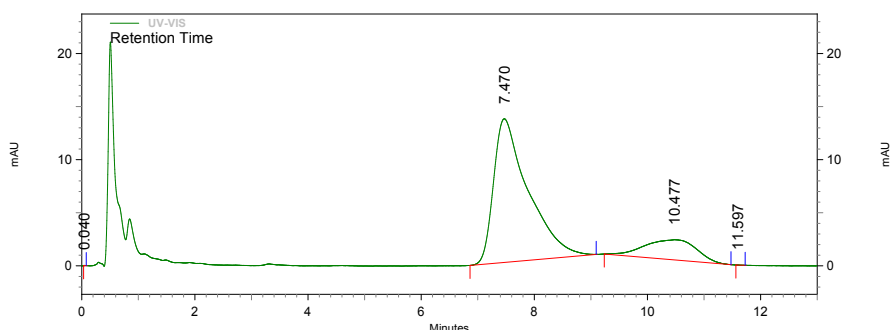
Sample ID: STD0.4 DEXAMETHSONE-0.04mg/ml Vial: 190 Injection Volume: 20



UV-VIS Results				
Name	Retention Time	Area	Area Percent	Integration Codes
	0.040	185	0.000	BI
	7.520	3077934	82.302	MM
	10.550	6618709	17.698	MM
Totals				
		9696828	100.000	

Analyst: LAB MANAGER

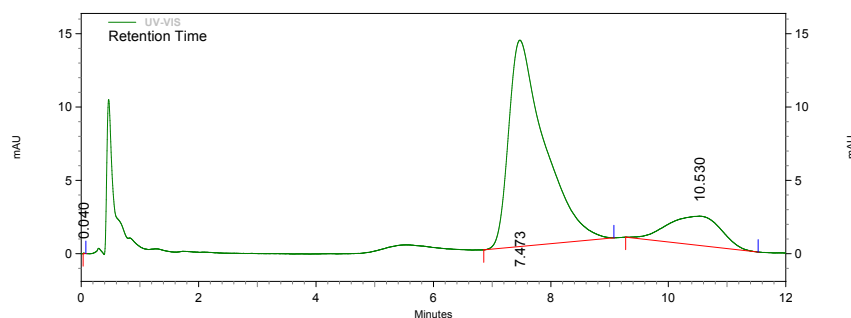
Sample ID: AARON DEXAMETHSONE-0.04mg/ml Vial: 195 Injection Volume: 20



UV-VIS Results				
Name	Retention Time	Area	Area Percent	Integration Codes
	0.040	188	0.007	BI
	7.470	2294125	82.677	MM
	10.477	480288	17.309	MM
	11.597	189	0.007	BI
Totals				
		2774790	100.000	

Analyst: LAB MANAGER

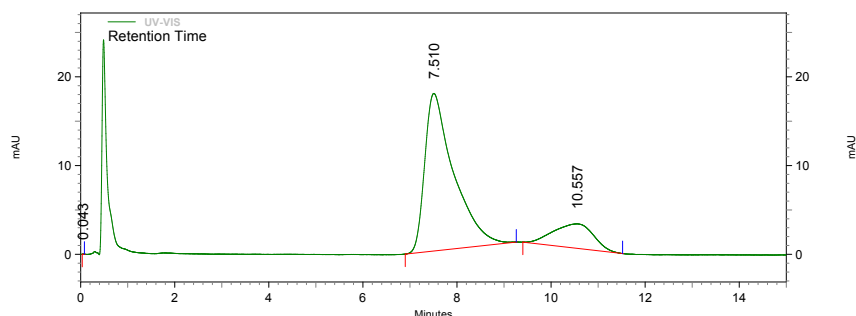
Sample ID: KRIS DEXAMETHSONE-0.04mg/ml Vial: 194 Injection Volume: 20



UV-VIS Results				
Name	Retention Time	Area	Area Percent	Integration Codes
	0.040	204	0.007	BI
	7.473	2388619	82.657	MM
	10.530	500974	17.336	MM
Totals				
		2889797	100.000	

Analyst: LAB MANAGER

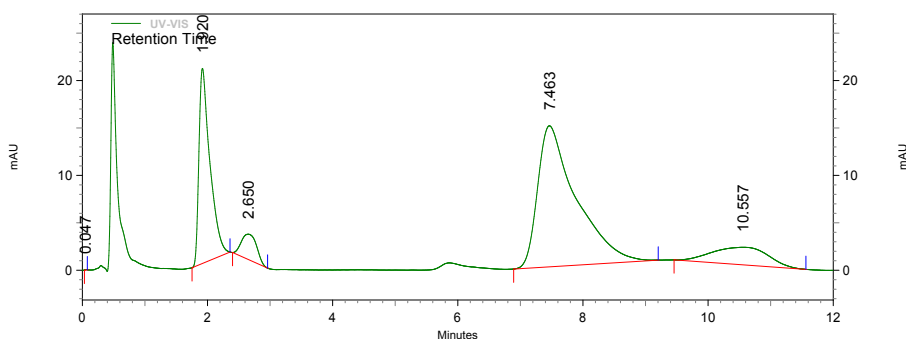
Sample ID: NKOYODEXAMETHSONE-0.04mg/ml Vial: 189 Injection Volume: 20



UV-VIS Results				
Name	Retention Time	Area	Area Percent	Integration Codes
	0.043	164	0.004	BI
	7.510	3038296	82.359	MM
	10.557	650610	17.636	MM
Totals				
		3689070	100.000	

Analyst: LAB MANAGER

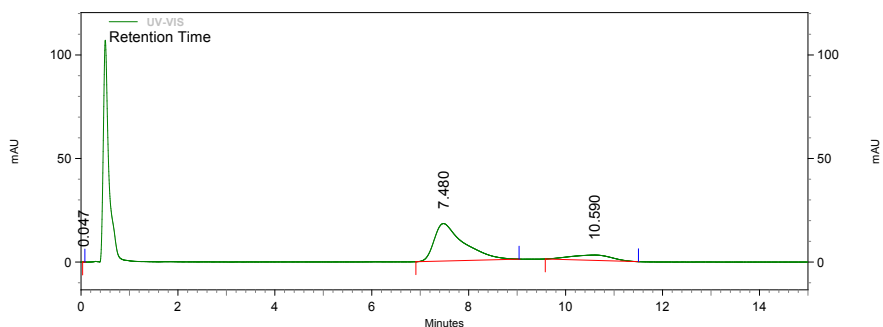
Sample ID: DEXAREL DEXAMETHSONE-0.04mg/ml Vial: 196 Injection Volume: 20



UV-VIS Results				
Name	Retention Time	Area	Area Percent	Integration Codes
	0.047	419	0.010	BI
	7.463	2537849	60.577	MM
	10.557	461496	11.016	MM
Totals				
		2999764	100.000	

Analyst: LAB MANAGER

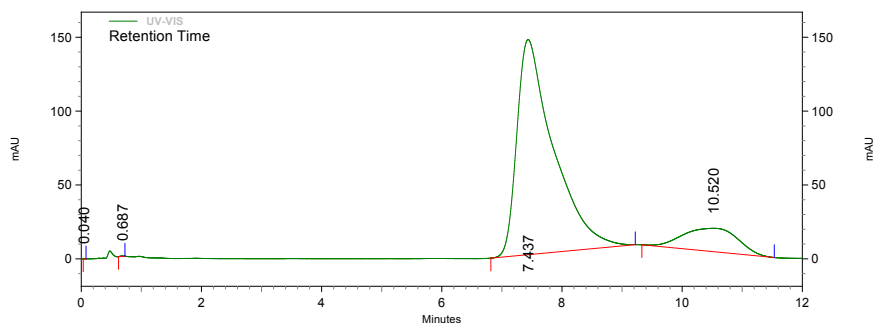
Sample ID: LOKEL DEXAMETHSONE-0.04mg/ml Vial: 188 Injection Volume: 20



UV-VIS Results				
Name	Retention Time	Area	Area Percent	Integration Codes
	0.047	189	0.005	BI
	7.480	3050263	83.497	MM
	10.590	602685	16.498	MM
Totals				
		3653137	100.000	

Analyst: LAB MANAGER

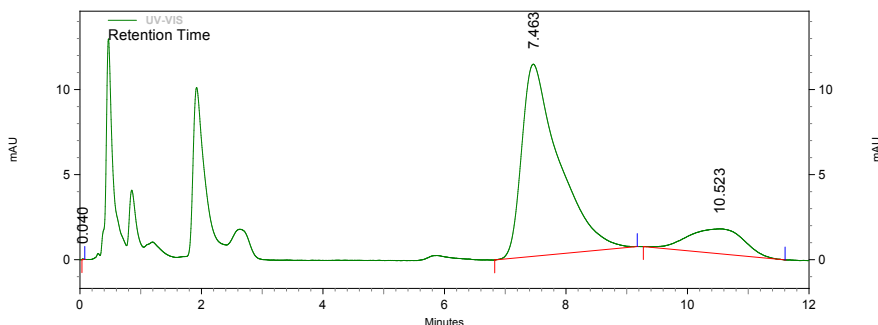
Sample ID: ANISUN DEXAMETHSONE-0.04mg/ml RPT Vial: 192 Injection Volume: 20



UV-VIS Results				
Name	Retention Time	Area	Area Percent	Integration Codes
	0.040	545	0.001	BI
	7.557	2217860	81.954	MM
	10.687	487808	18.025	MM
Totals				
		2706213	100.000	

Analyst: LAB MANAGER

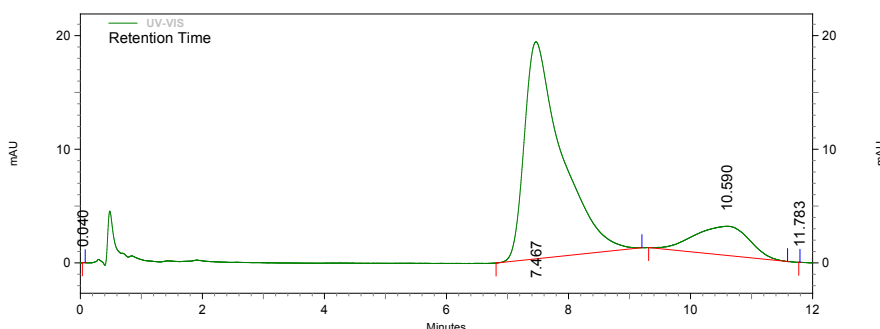
Sample ID: CHAZMAX DEXAMETHSONE-0.04mg/ml Vial: 193 Injection Volume: 20



UV-VIS Results				
Name	Retention Time	Area	Area Percent	Integration Codes
	0.040	203	0.009	BI
	7.463	1932916	83.872	MM
	10.523	371485	16.119	MM
Totals				
		2304604	100.000	

Analyst: LAB MANAGER

Sample ID: DEXA DEXAMETHSONE-0.04mg/ml Vial: 197 Injection Volume: 20



UV-VIS Results				
Name	Retention Time	Area	Area Percent	Integration Codes
	0.040	231	0.006	BI
	7.467	3247323	83.609	MM
	10.590	636401	16.385	MM
	11.783	5	0.000	BI
Totals				
		3883960	100.000	

Calculation of percentage content of the samples using HPLC method of analysis

$$\% \text{ Content} = \frac{\text{peak area of sample}}{\text{Peak area of standard}} \times 100\%$$

STANDARD (peak area = 3077934.)

$$\% \text{ Content} = \frac{3077934}{3077934} \times 100\% = 100\%$$

AARON'S (peak area = 2294125.)

$$\% \text{ Content} = \frac{2294125}{3077934} \times 100\% = 74.5\%$$

KRIS (peak area = 2388619.)

$$\% \text{ Content} = \frac{2388619}{3077934} \times 100\% = 77.6\%$$

NKOYO (peak area = 3038296.)

$$\% \text{ Content} = \frac{3038296}{3077934} \times 100\% = 98.7\%$$

DEXAREL (peak area = 2537849.)

$$\% \text{ Content} = \frac{2537849}{3077934} \times 100\% = 82.5\%$$

LOKEL'S (peak area = 3050263.)

$$\% \text{ Content} = \frac{3050263}{3077934} \times 100\% = 99.1\%$$

ANISUN (peak area = 2217860.)

$$\% \text{ Content} = \frac{2217860}{3077934} \times 100\% = 72.1\%$$

CHAZMAX (peak area = 1932916.)

$$\% \text{ Content} = \frac{1932916}{3077934} \times 100\% = 62.8\%$$

DEXA (peak area = 3247323.)

$$\% \text{ Content} = \frac{3247323}{3077934} \times 100\% = 105.5\%$$

TABLE 4: SHOWING THE PERCENTAGE CONTENT OF THE SAMPLE USING HPLC METHOD OF ANALYSIS.

BRAND	PEAK AREA	% CONTENT
STANDARD	3077934	100
AARON'S	2294125	74.5
KRIS	2388619	77.6
NKOYO	3038296	98.7
DEXAREL	2537849	82.5
LOKEL'S	3050263	99.1
ANISUN	2217860	72.1
CHAZMAX	1932916	62.8
DEXA	3247323	105.5

Calculations of milligram (mg) content of the samples using UV- spectrophotometry at a wave length of 238nm.

$$\text{Mg Content} = \frac{\% \text{ content of sample}}{100} \times \text{stated claim in mg}$$

STANDARD (% content = 100 %.)

$$\text{Mg Content} = \frac{100\%}{100} \times 0.5\text{mg} = 0.5\text{mg}$$

AARON'S (% content = 116.2 %.)

$$\text{Mg Content} = \frac{116.2\%}{100} \times 0.5\text{mg} = 0.6\text{mg}$$

KRIS (% content = 79.6 %.)

$$\text{Mg Content} = \frac{79.6\%}{100} \times 0.5\text{mg} = 0.4\text{mg}$$

NKOYO (% content = 98.1 %.)

$$\text{Mg Content} = \frac{98.1\%}{100} \times 0.5\text{mg} = 0.5\text{mg}$$

DEXAREL (% content = 85.7 %.)

$$\text{Mg Content} = \frac{85.7\%}{100} \times 0.5\text{mg} = 0.4\text{mg}$$

LOKEL'S (% content = 99.2 %.)

$$\text{Mg Content} = \frac{99.2\%}{100} \times 0.5\text{mg} = 0.5\text{mg}$$

ANISUN (% content = 79.6 %.)

$$\text{Mg Content} = \frac{79.6\%}{100} \times 0.5\text{mg} = 0.4\text{mg}$$

CHAZMAX (% content = 100.1 %.)

$$\text{Mg Content} = \frac{86.5\%}{100} \times 0.5\text{mg} = 0.4\text{mg}$$

DEXA (% content = 95.8 %.)

$$\text{Mg Content} = \frac{95.8\%}{100} \times 0.5\text{mg} = 0.5\text{mg}$$

TABLE 5: SHOWING THE MILLIGRAM (mg) CONTENT OF THE SAMPLES USING UV-SPECTROPHOTOMETRY AT A WAVE LENGTH OF 238nm.

BRAND	% CONTENT	Mg CONTENT
STANDARD	100	0.5
AARON'S	116.2	0.6
KRIS	79.6	0.4
NKOYO	98.1	0.5
DEXAREL	85.7	0.4
LOKEL'S	99.2	0.5
ANISUN	79.6	0.4
CHAZMAX	86.5	0.4
DEXA	95.8	0.5

Calculation of milligram (mg) content of the samples using HPLC method of analysis.

$$\text{Mg Content} = \frac{\% \text{ content of sample}}{100} \times \text{stated claim in mg}$$

STANDARD (% content = 100 %.)

$$\text{Mg Content} = \frac{100\%}{100} \times 0.5\text{mg} = 0.5\text{mg}$$

AARON'S (% content = 74.5 %.)

$$\text{Mg Content} = \frac{74.5\%}{100} \times 0.5\text{mg} = 0.4\text{mg}$$

KRIS (% content = 77.6 %.)

$$\text{Mg Content} = \frac{77.6\%}{100} \times 0.5\text{mg} = 0.4\text{mg}$$

NKOYO (% content = 98.7 %.)

$$\text{Mg Content} = \frac{98.7\%}{100} \times 0.5\text{mg} = 0.5\text{mg}$$

DEXAREL (% content = 82.5 %.)

$$\text{Mg Content} = \frac{82.5\%}{100} \times 0.5\text{mg} = 0.4\text{mg}$$

LOKEL'S (% content = 99.1 %.)

$$\text{Mg Content} = \frac{99.1\%}{100} \times 0.5\text{mg} = 0.5\text{mg}$$

ANISUN (% content = 72.1 %.)

$$\text{Mg Content} = \frac{72.1\%}{100} \times 0.5\text{mg} = 0.4\text{mg}$$

CHAZMAX (% content = 62.8 %.)

$$\text{Mg Content} = \frac{62.8\%}{100} \times 0.5\text{mg} = 0.3\text{mg}$$

DEXA (% content = 105.5 %.)

$$\text{Mg Content} = \frac{105.5\%}{100} \times 0.5\text{mg} = 0.5\text{mg}$$

TABLE 6: SHOWING THE MILIGRAM (mg) CONTENT OF THE SAMPLE USING HPLC METHOD OF ANALYSIS

BRAND	% CONTENT	Mg CONTENT
STANDARD	100	0.5
AARON'S	74.5	0.4
KRIS	77.6	0.4
NKOYO	98.7	0.5
DEXAREL	82.5	0.4
LOKEL'S	99.1	0.5
ANISUN	72.1	0.4
CHAZMAX	62.8	0.3
DEXA	105.5	0.5

DISCUSSION

This study was carried out to quantify the various brands of dexamethasone tablets marketed in Maiduguri metropolis and to ascertain whether they are of the standard specified in the official compendiums.

From the result obtained, it is observed that using the UV-spectrophotometry, three of the samples (NKOYO, LOKEL'S and DEXA) out of the eight have a percentage content of 98.1%, 99.2% and 95.8% respectively which falls within the limit stated by the BP (90-110%) for uniformity of content. Four of the samples (KRIS, DEXAREL, ANISUN and CHAZMAX) had a percentage content falling below the standard limit stated by the BP. The other sample (ARRON'S) had percentage content (116.2%) above the BP limit.

The HPLC method of analysis gave results that correspond to that obtained by the UV-spectrophotometric analysis. The three samples that passed in the UV-spectrophotometry also passed in the HPLC analysis, while the remaining five

samples that failed in the UV-spectrophotometry also failed in the HPLC analysis, indicating a validation of the result obtained.

These deviations from the BP stated limit could result from inappropriate formulation, poor storage facilities, adulteration either during the production process or after production.

The environmental condition of Maiduguri could probably result in the deterioration of pharmaceutical products (drugs) owing to its temperature (heat), oxygen, light (photochemical decomposition), changes in pH and the unavailability of good and adequate storage facilities.

Product related factors that could result in the deterioration of drugs includes; the active ingredient and excipients, the dosage form and its composition, method of production as well as the nature & property of the packaging materials.

CONCLUSION

Out of the eight different samples of dexamethasone tablet analysed, three NKOYO, LOKEL'S and DEXA passed the laid down specification while the other five samples KRIS,

DEXAREL, ANISUN, CHAZMAX and ARRON'S failed probably due to the reasons stated above.

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