An Overview on Various Analytical Methods for Estimation of Atenolol and Amiodarone from its Bulk and Pharmaceutical Dosage Forms

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ABSTRACT
The main objective of this review is to unify and interpret widely scattered information of reported studies on potential, reliable and efficient analytical methodologies which can estimate Atenolol and Amiodarone separately. The information and suggested outlined below may facilitate and guide further needed studies to optimize the use of analytical techniques like High Performance Liquid Chromatography (HPLC), Bioanalytical Methods, UV Spectroscopy, Stability indicating RP-HPLC methods etc. for determination of Atenolol and Amiodarone in formulation. From the reviewed literature it is obvious that HPLC is a commonly available method of testing in pharmaceutical laboratory so this method should be of choice for complete determination of Atenolol and Amiodarone. Selection of analytical methods is determined by several factors such as speed, convenience, specificity, accuracy, precision, sensitivity, selectivity, cost, availability of instruments, technical expertise and the number of samples to be analyzed.

Keywords: Atenolol, Amiodarone, Analytical estimation, HPLC, UV

INTRODUCTION
Atenolol is a beta blocker medication primarily used to treat high blood pressure and heart-associated chest pain. Atenolol, however, does not seem to improve mortality in those with high blood pressure (AHFS, 2018; Tomiyama H and Yamashina A, 2014; DiNicolantonio JJ, et al., 2015). Other uses include the prevention of migraines and treatment of certain irregular heartbeats. It is taken by mouth or by injection into a vein. It can also be used with other blood pressure medications (British National Formulary, 2018).

Common side effects include feeling tired, heart failure, dizziness, depression, and shortness of breath. Other serious side effects include bronchospasm. Use is not recommended during pregnancy and alternative drugs are preferred when breastfeeding. It works by blocking β1-adrenergic receptors in the heart, thus decreasing the heart rate and workload.

Atenolol was patented in 1969 and approved for medical use in 1975. It is available as a generic medication. In 2018, it was the 42nd most commonly prescribed medication in the United States, with more than 18 million prescriptions (Beard Jr EL, 2001; Ali MU, et al., 2018; Florey K, 1981; Akiful HM, et al., 2012; Godge RK, et al., 2017) (Figure 1).

Amiodarone is an antiarrhythmic medication used to treat and prevent a number of types of irregular heartbeats (Beard Jr EL, 2001). This includes Ventricular Tachycardia (VT), Ventricular Fibrillation (VF), and wide complex tachycardia, as well as atrial fibrillation and paroxysmal supraventricular tachycardia (Ali MU, et al., 2018). Evidence in cardiac arrest, however, is poor. It can be given by mouth, intravenously, or intrasoosseously. When used by mouth, it can take a few weeks for effects to begin. Common side effects include feeling tired, tremor, nausea, and constipation (Florey K, 1981). As amiodarone can have serious side effects, it is mainly recommended only for significant ventricular arrhythmias. Serious side effects include lung toxicity such as interstitial pneumonitis, liver problems, heart arrhythmias, vision problems, thyroid problems, and death. If taken during pregnancy or breastfeeding it can cause problems in the fetus. It is a class III antiarrhythmic medication. It works partly by increasing the time before a heart cell can contract again.

Amiodarone was first made in 1961 and came into medical use in 1962 for chest pain believed to be related to the heart. It was pulled from the market in 1967 due to side effects. In 1974 it was found to be useful for arrhythmias and reintroduced. It is on the World Health Organization’s List of Essential Medi-
The robustness of an analytical technique is a measure of its ability to remain unaffected by minor yet deliberate changes in system parameters, and it indicates its efficiency during regular use.

**Accuracy in the middle:**

**Precisely in the middle of the day:** It is carried out by making another researcher analyse the data on a different day to ensure that the findings are repeatable. Samples prepared in the same way that the Repeatability parameter samples were (6 Samples prepared).

**Criteria for acceptance:** For test results, the percent RSD of 6 samples NMT 2.0% was used.

NMT 2.0% for test results, percent RSD of total 12 samples.

(6 of Repeatability and 6 of Intermediate precision)

**Robustness:** The robustness of an analytical technique is a measure of its ability to remain unaffected by substantial changes in system parameters, and it indicates its efficiency during regular use.

**Detection:**

**Limit Of Detection (LOD):** Under the specified experimental conditions, the lowest concentration of the analyte in the sample that the system can detect but not necessarily quantify simply means that the sample is below or above a certain threshold. Limits are defined in percentages or parts per million. The detection limit will be calculated not only by the measurement technique, but also by the type of instrument used.

\[
S/N = \frac{S}{N} = \frac{2}{1} \text{ or } \frac{3}{1}
\]

Where, \(S\) = Signal, \(N\) = Noise

It may be calculated based on the Standard Deviation (SD) of the response and slope of the curve(S).

\[
LOD = 3.3 \times (SD)/S
\]

Where, SD = Standard deviation, S = Slope

**Limit Of Quantitation (LOQ):** The lowest amount of analyte in a sample that can be calculated with reasonable precision and accuracy under the specified experimental conditions is known as the limit of quantitation (LOQ). It is expressed as the percentage of analyte in the sample (e.g., parts per billion). The S/N ratio should not be less than 10 and the RSD should be less than 3%.

\[
S/N = 10/1
\]

Where \(S\) = Signal \(N\) = Noise

It may be calculated based on the Standard Deviation (SD) of the response and slope of the curve(S).

\[
LOQ = 10 \times (SD)/S
\]

Where, SD = Standard deviation, S = Slope

**Experimental work**

Literature survey revealed that was determined by UV-visible spectroscopy and HPLC. In the current work, the authors have proposed a simple, specific, valid and robust RP-HPLC method for the estimation of Atenolol and amiodarone in pharmaceutical active substance form (Tables 1 and 2).
Table 1: Analytical methods used for the estimation of Atenolol from bulk and formulations

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Name of author</th>
<th>Name of journal</th>
<th>Title of article</th>
<th>Analytical conditions</th>
</tr>
</thead>
<tbody>
<tr>
<td>UV Spectrophotometric</td>
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</table>
λ max-549 nm  
Beer-Lambert's limits (µg/mL)- 2-10  
linear regression equation-  
Y=0.0572 C+0.0033  
correlation coefficient-0.9990  
% RSD-0.319  
% Recovery-99.5%  
LOD-5.88 µg/mL  
LOQ-17.83 µg/mL |
| Turbidimetric analysis | | | | |
| 1 | Al-Awadie NS and Khudhair AF (Al-Awadie NS and Khudhair AF, 2014) | Iraqi Journal of Science | Determination of Atenolol in pharmaceutical formulations by continuous flow injection analysis via turbidimetric (T180º) and scattered light effect at two opposite position (2N90º) using Ayah 4SW-3D-T180-2N90-Solar-CFI Analyzer | Turbidimetric: T180º  
Scattered light effect at two opposite position (2N90º).  
Incident light in namely +90º and -90º  
Linearity of Atenolol is ranged from (0.1-11) mmol. L⁻¹  
Correlation coefficient-0.938  
LOD-0.05 mmol. L⁻¹ |
| Kinetic method | | | | |
103 [Unknown-1] moldm⁻³ = 3.75(calculated): 3.75(actual)  
103 [Unknown-2] moldm⁻³ = 6.26 ± 0.01(calculated): 6.25(actual) |
| Bioanalytical methods | | | | |
M.P.- methanol=water (50:50, v/v)  
λ max-549 nm  
Beer-Lambert's limits-  
5-150 ng/mL  
correlation coefficient-0.9990  
% RSD-0.319  
% Recovery-98.4%  
LOD-1.5 ng/mL  
LOQ-5 ng/mL |
| Stability-Indicating HPLC method | | | | |
M.P.-acetonitrile: methanol:0.02 M phosphate buffer, pH 5 (20:20:60)  
Flow rate-1 ml/min  
λ max-226 nm  
Beer-Lambert's limits-  
0.05-10 µg/ml  
Correlation coefficient-1  
% Recovery-100.4%  
LOD-0.01 µg/mL  
LOQ-0.03 µg/mL  
stability-indicating capability-acid and base media |
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UV-HPLC

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<tbody>
<tr>
<td>1</td>
<td>Goebel K and Rolim CM (Goebel K and Rolim CM, 2007)</td>
<td>Latin American Journal of Pharmacy</td>
<td>Validation of UV Spectrophotometric and HPLC Methods for Quantitative Determination of Atenolol in Pharmaceutical Preparations.</td>
<td>Column-Purospher RP-18 (250 mm × 4.6 mm, 5 μm) Solvent-10 mM ammonium acetate buffer (pH 7.0) and acetonitrile (80:20 v/v) λ max-275 nm Beer-Lambert’s limits (μg/mL)- 2-10 Correlation coefficient-0.9990 % Recovery-98.5%</td>
</tr>
<tr>
<td>2</td>
<td>Kumar N, et al. (Kumar N, et al., 2010)</td>
<td>E-Journal of Chemistry</td>
<td>Estimation of Atenolol by Reverse Phase High Performance Liquid Chromatography</td>
<td>Column-ODS and dimensions of column was 25 mm × 4.6 mm M.P-phosphate buffer and acetonitrile (53:47 v/v) λ max-230 nm Flow rate-2.1 mL/min Beer-Lambert’s limits- 5-150 ng/mL correlation coefficient-0.9990 % RSD-0.6 % Recovery-99.6% LOD-510 ng/mL LOQ-120 ng/mL</td>
</tr>
<tr>
<td>3</td>
<td>Kori S, et al. (Kori S, et al., 2013)</td>
<td>International Journal of Science and Research</td>
<td>Method Development and Validation of Atenolol Drug by Spectrophotometric and HPLC Technique in Forensic Application</td>
<td>Column-RPC18 column λ max-226 nm Flow rate-2.1 mL/min Beer-Lambert’s limits- 25-50 μg/mL correlation coefficient-0.9990 % RSD-0.5 % Recovery-99.5% LOD-2.00 μg/mL LOQ-6.3 μg/mL</td>
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Reverse Phase High Performance Liquid Chromatography

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</thead>
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<tr>
<td>1</td>
<td>Al-Rimawi F (Al-Rimawi F, 2010)</td>
<td>Pharmaceutica Analytica Acta</td>
<td>Validation of an HPLC-UV Method for the Determination of Amiodarone Impurities in Tablet Formulations</td>
<td>Column- C18 column λ max-240 nm Mob. Phase-buffer solution pH 5.0, methanol, and acetonitrile (30:30:40, v/v/v) Beer-Lambert’s limits (μg/mL)- 0.005-0.015 Correlation coefficient-0.9990 % Recovery-99.7% LOD-0.0005 μg/mL LOQ-0.0002 μg/mL</td>
</tr>
</tbody>
</table>

Table 2: Analytical methods used for the estimation of Amiodarone from bulk and formulations
### Bioanalytical methods

| | Rodrigues M, et al., 2013 | Journal of Chromatographic Science | Mob. Phase- phosphate buffer (50 mM) with 0.1% formic acid (pH 3.1)-methanol-acetonitrile (45:5:50, v/v/v) | Flow rate-1.3 mL/min
| | | | Beer-Lambert’s limits (µg/mL)- 0.1-15 | 0.995 correlation coefficient-0.995
| | | | % Recovery-97.7 %
| Bioanalytical methods | 2 Jun AS and Brocks DR (Jun AS and Brocks DR, 2001) | High performance liquid chromatographic assay of amiodarone in rat plasma. | Column-C8 analytical column | λ max-242 nm
| | | | Mob. Phase- buffer solution pH 5.0, methanol, and acetonitrile (30:30:40, v/v/v) | 0.998 correlation coefficient-0.9998
| | | | Correlation coefficient-0.9998 | % Recovery-75-82%
| | | | LOD-0.035 µg/mL | LOQ-0.035 µg/mL

### RP-HPLC

| | Asian Journal of Pharmaceutical Analysis and Medicinal Chemistry | | Mob. Phase- Acetonitrile: 0.5%Triethylamine Buffer pH to 6.5 with orthophosphoric acid (75:25) | Flow rate-2.0 mL/min
| | | | Correlation coefficient-0.9990 | % Recovery-99.7-100.1%
| | | | % Recovery-75-82% | LOD-0.035 µg/mL
| | | | LOQ-0.035 µg/mL |
| | Journal of Liquid Chromatography and Related Technologies | | Flow rate-1.5 mL/min |
| | | | Mob. Phase- methanol, water, and acetic acid in a 95:4:1 | Linear equation-y ¼ 27683 x b 42192 Correlation coefficient-0.94
| | | | Correlation coefficient-0.94 | % Recovery-99.7%
| | | | LOD-3.12 µg/mL | LOQ-0.936 µg/mL
| | | | % Recovery-99.7% |
| | World Journal of Pharmaceutical Research | | Flow rate-1.0 mL/min |
| | | | Mob. Phase- Acetonitrile: Water (80:20) | Degradation Study-acid hydrolysis (1 M HCl at 60°C for 3 hrs),
| | | | Degradation Study-oxidation (6% H₂O₂ at 60°C for 3 hrs) | basic hydrolysis (1 M NaOH at 60°C for 3 hrs)
| | | | oxidation (6% H₂O₂ at 60°C for 3 hrs) |

### Stability indicating RP-HPLC method

| Stability indicating RP-HPLC method | 1 Mallu UR, et al. (Mallu UR, et al., 2010) | Method Development of stability indicating HPLC method for the determination of Amiodarone Hydrochloride in pharmaceutical dosage form | Column-Novapak C8 column 3.9*150 mm-particle size of 4 mm | λ max-240 nm
| | Drug Invention Today | | Flow rate-1.0 mL/min |
| | | | Mob. Phase-acetate buffer-Acetonitrile (15:85 v/v) | Linearity range-12.5 to 75 µg/mL
| | | | Correlation coefficient-0.999 | % Recovery-99.24 %
CONCLUSION
Presented work is focused on the use of different analytical methods like High Performance Liquid Chromatography (HPLC), Bioanalytical Methods, UV Spectroscopy, Stability indicating RP-HPLC methods etc. for determination of Atenolol and Amiodarone in formulation as well as in API. From the reviewed literature it is obvious that HPLC is a commonly available method of testing in pharmaceutical laboratory so this method should be of choice for complete determination of Atenolol and Amiodarone. No one analytical methods are available in market for the simultaneous estimation of the Atenolol and Amiodarone in Pharmaceutical dosage form and bulk drugs.

REFERENCES
1. AHFS. Atenolol Monograph for Professionals”. Drugs. 2018.