Electronic Supplementary Material (ESI) for Analytical Methods.

Electronic Supplementary Information
For

A novel HPLC-DAD method for simultaneous determination of alfuzosin and solifenacin along with their official impurities induced *via* a stress stability study; investigation of their degradation kinetics

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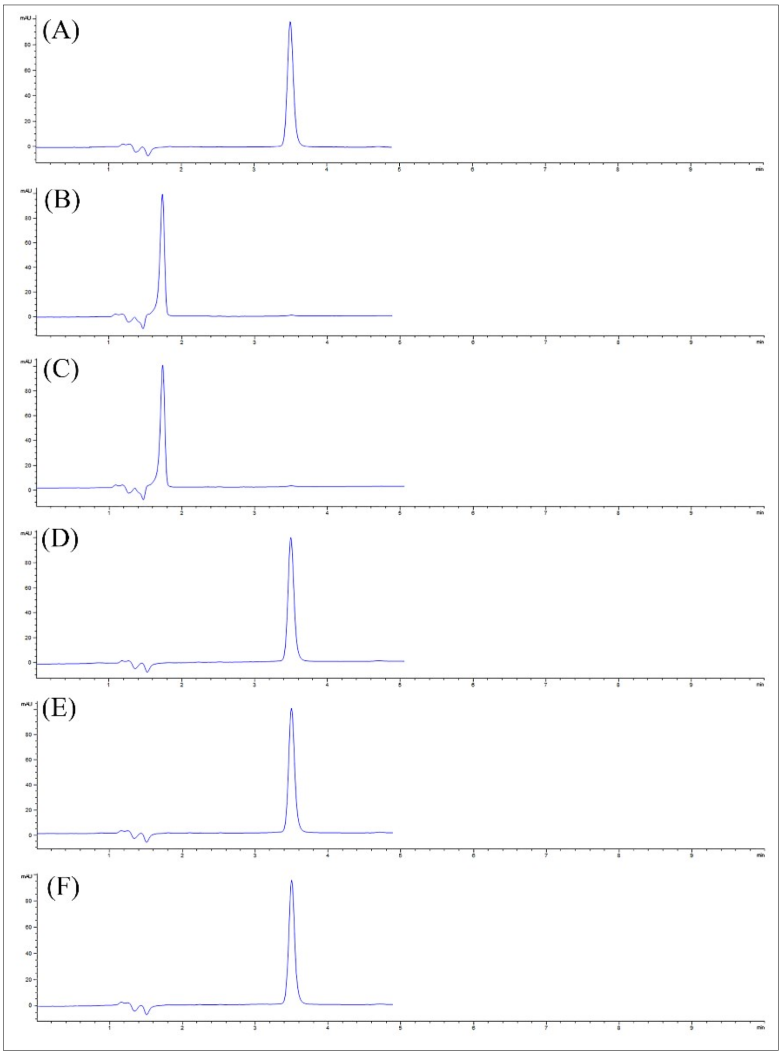


Fig. S1. HPLC chromatograms of the stability of alfuzosin HCl [retention time (t_R) = 3.8 min] in different stress conditions, showing the disappearance of its peak after acidic or alkaline hydrolysis and the appearance of a new peak corresponding to ALF imp-D at t_R = 1.7 min where (A) intact drug; (B) acidic hydrolysis in 0.5 M HCl; (C) basic hydrolysis in 0.5 M NaOH; (D) oxidative degradation in 3% hydrogen peroxide; (E) photolytic degradation; (F) thermal degradation in 100°C oven.

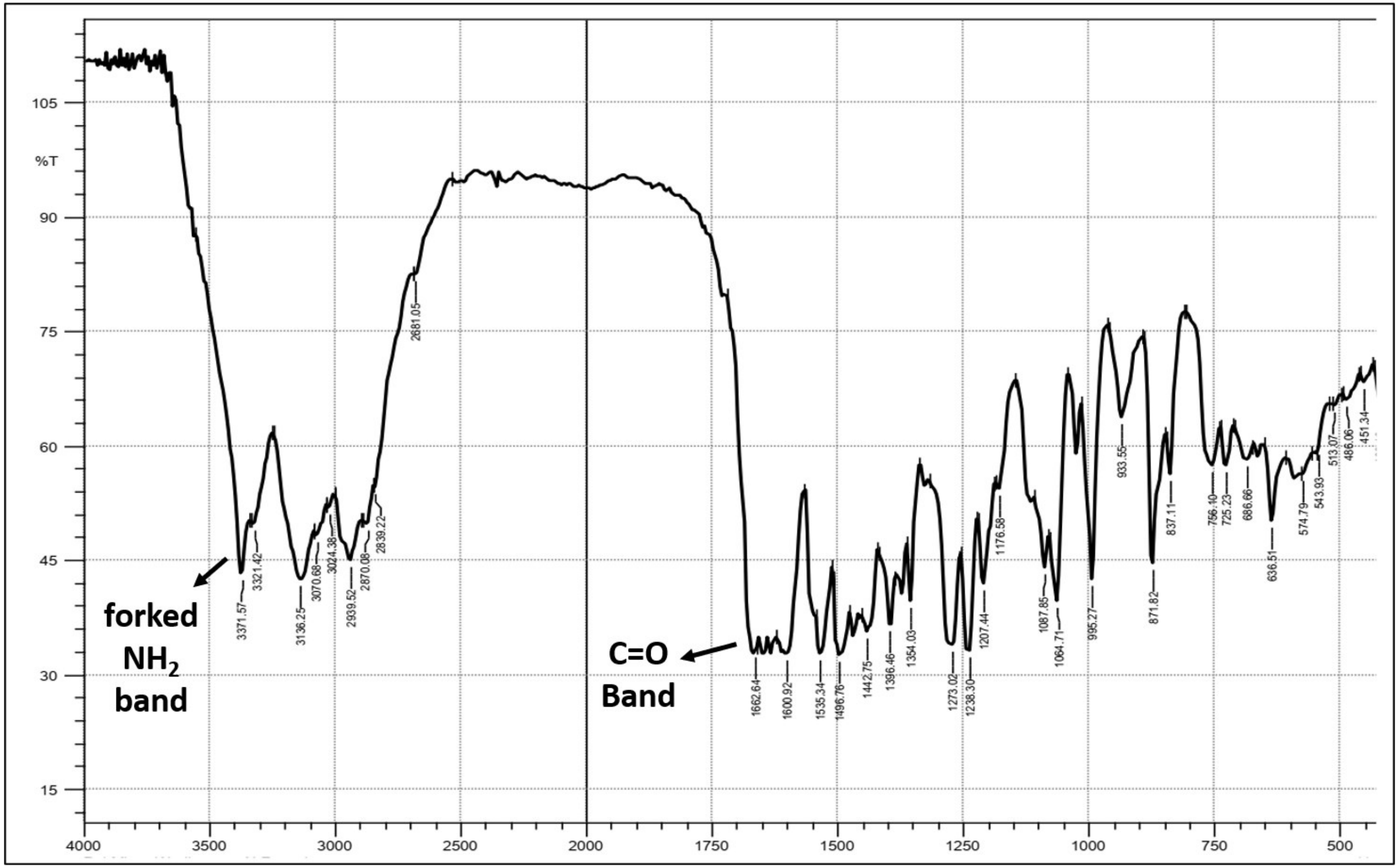


Fig. S2. (A) IR spectrum of ALF intact drug.

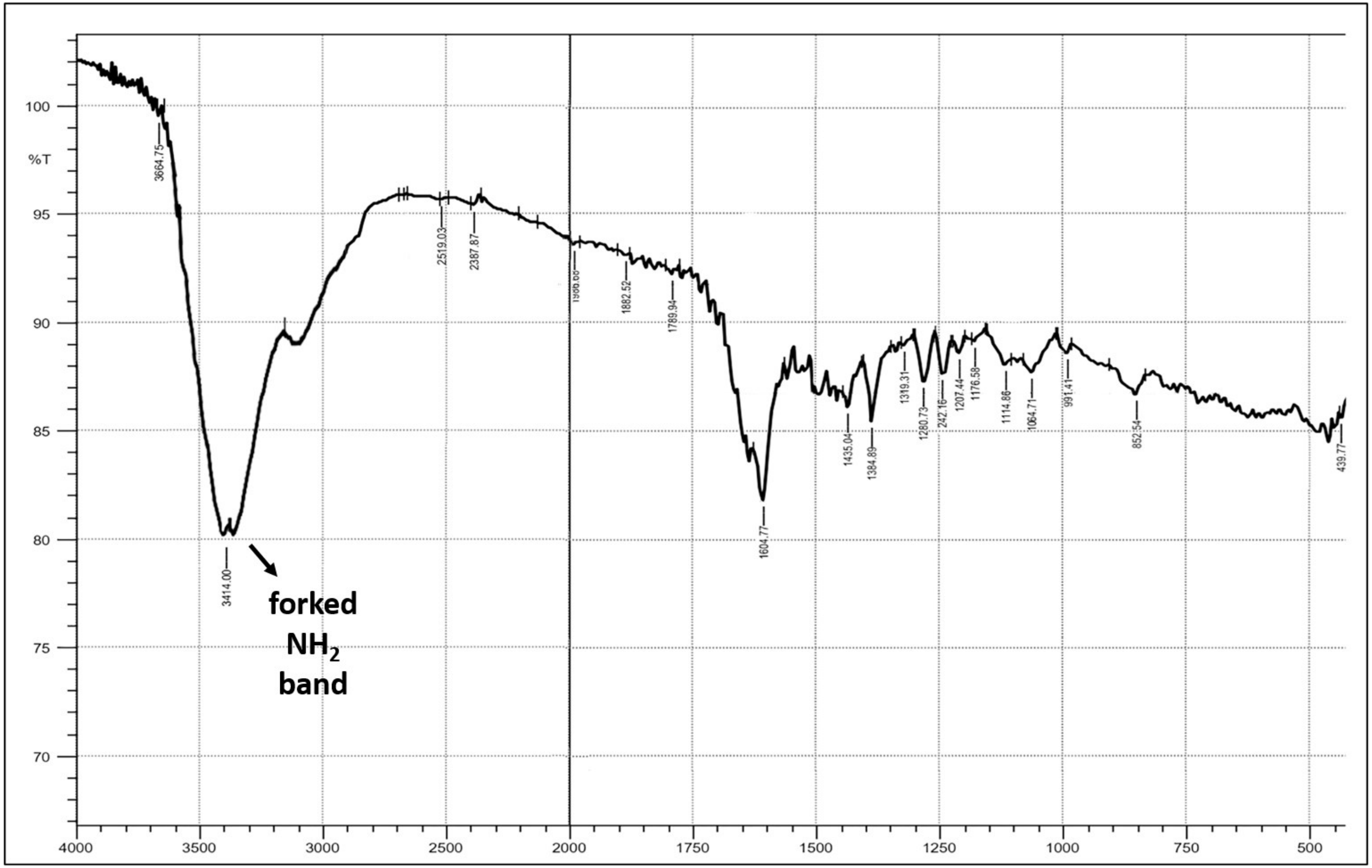


Fig. S2. (B) IR spectrum of ALF acidic induced degradation product.

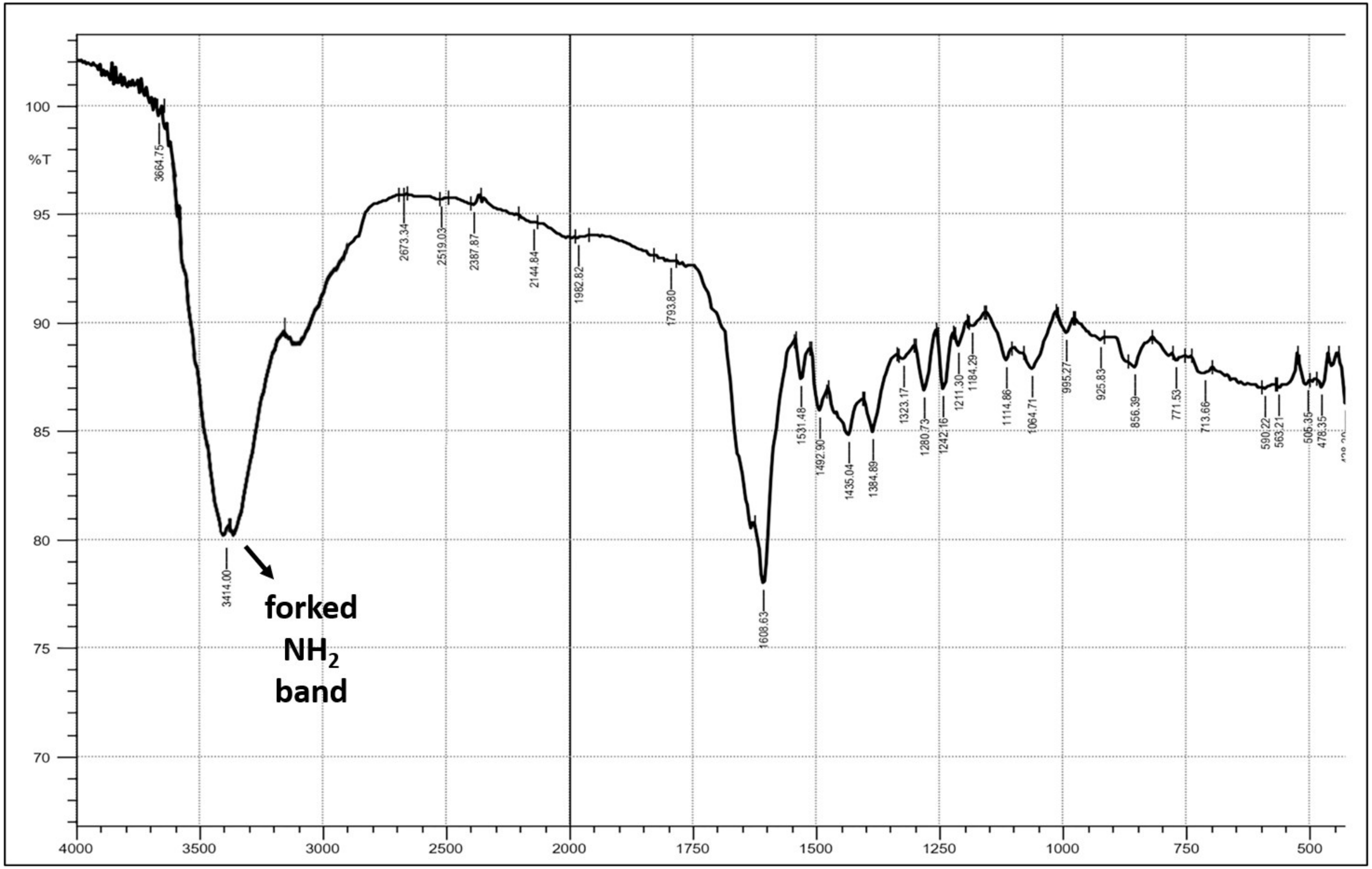


Fig. S2. (C) IR spectrum of ALF basic induced degradation product.

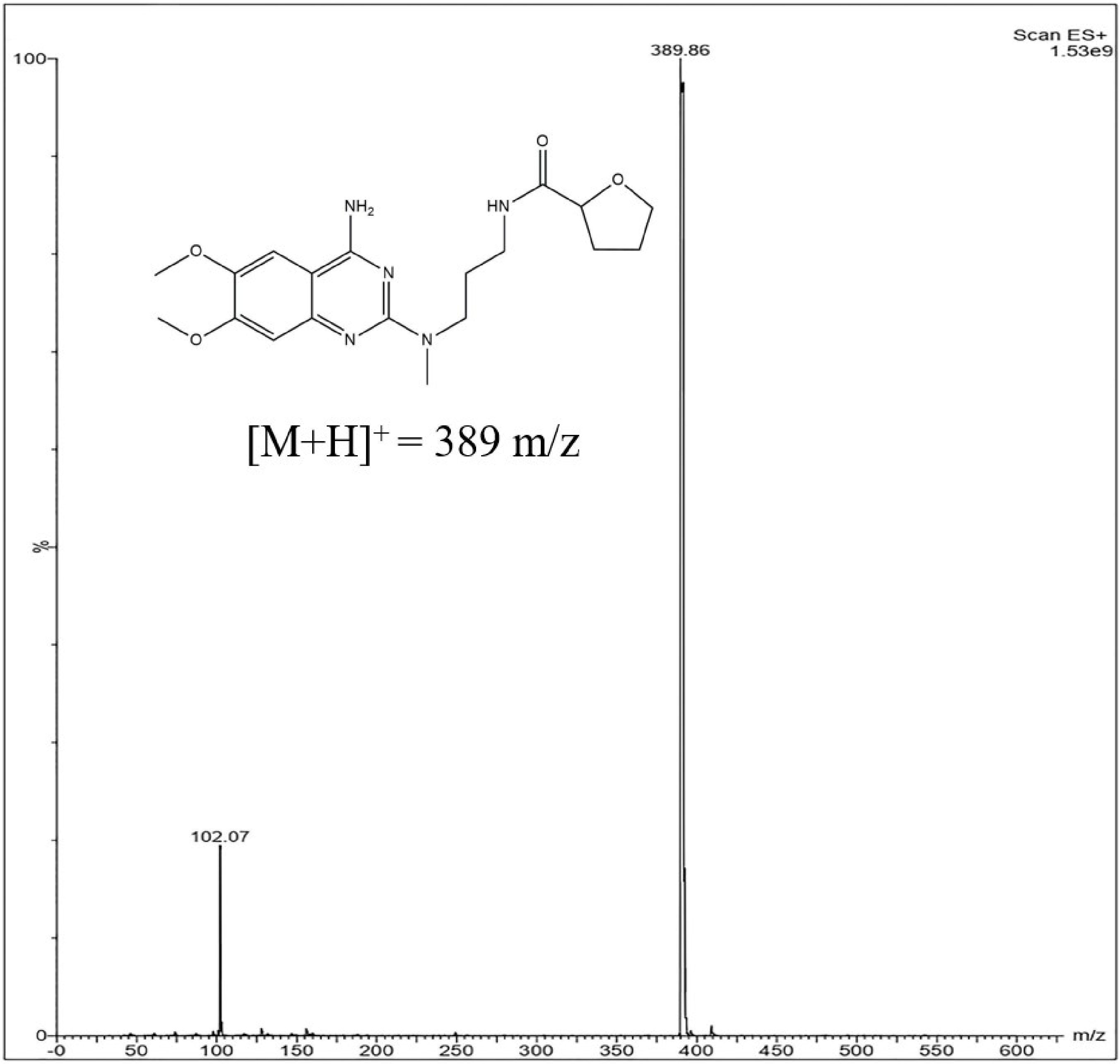


Fig. S3. (A) The mass spectrum showing molecular ion peak at 389 m/z for ALF intact drug.

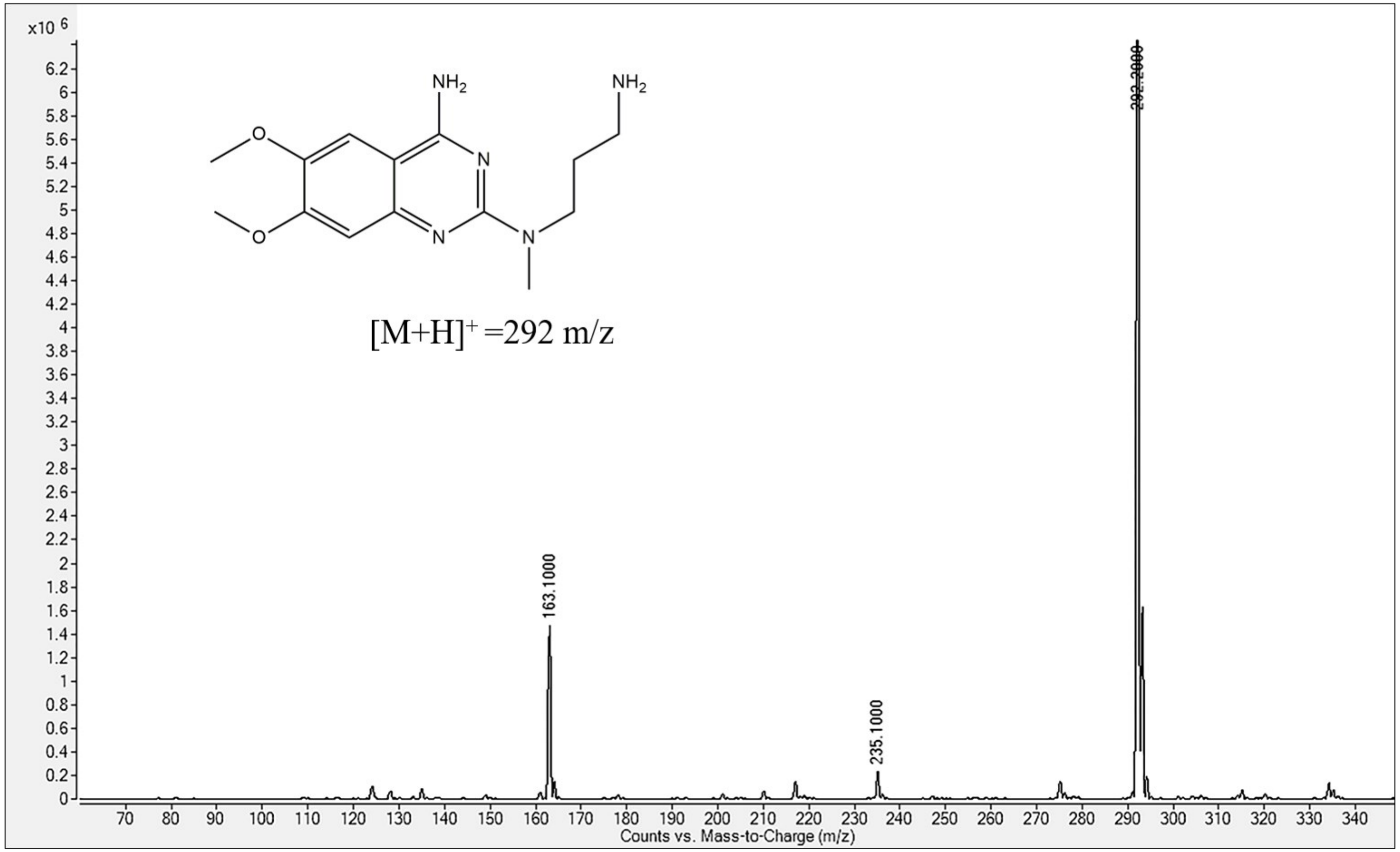


Fig. S3. (B) The mass spectrum showing molecular ion peak at 292 m/z for ALF acidic induced degradation product; ALF imp-D.

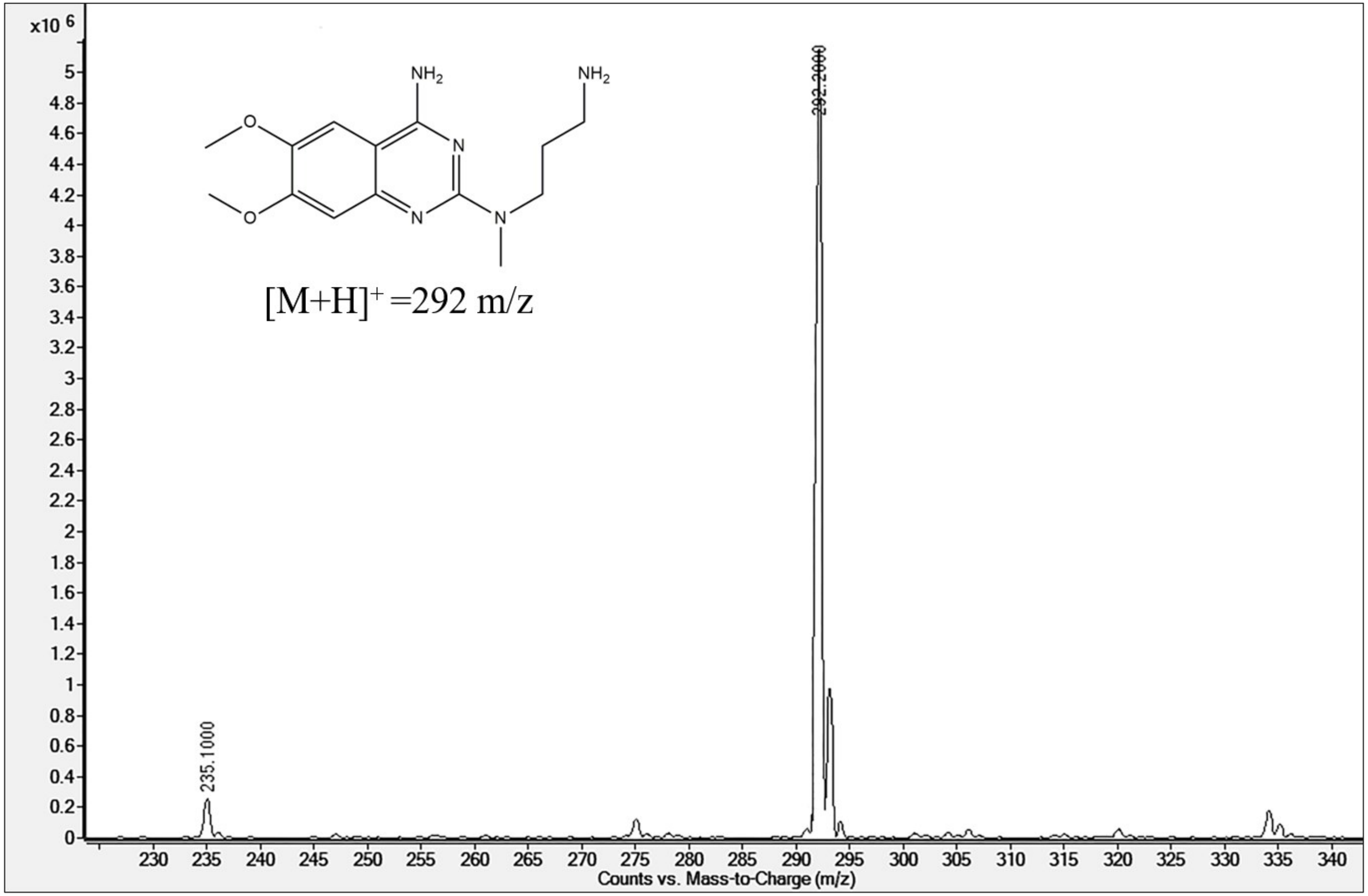


Fig. S3. (C) The mass spectrum showing molecular ion peak at 292 m/z for ALF basic induced degradation product; ALF imp-D.

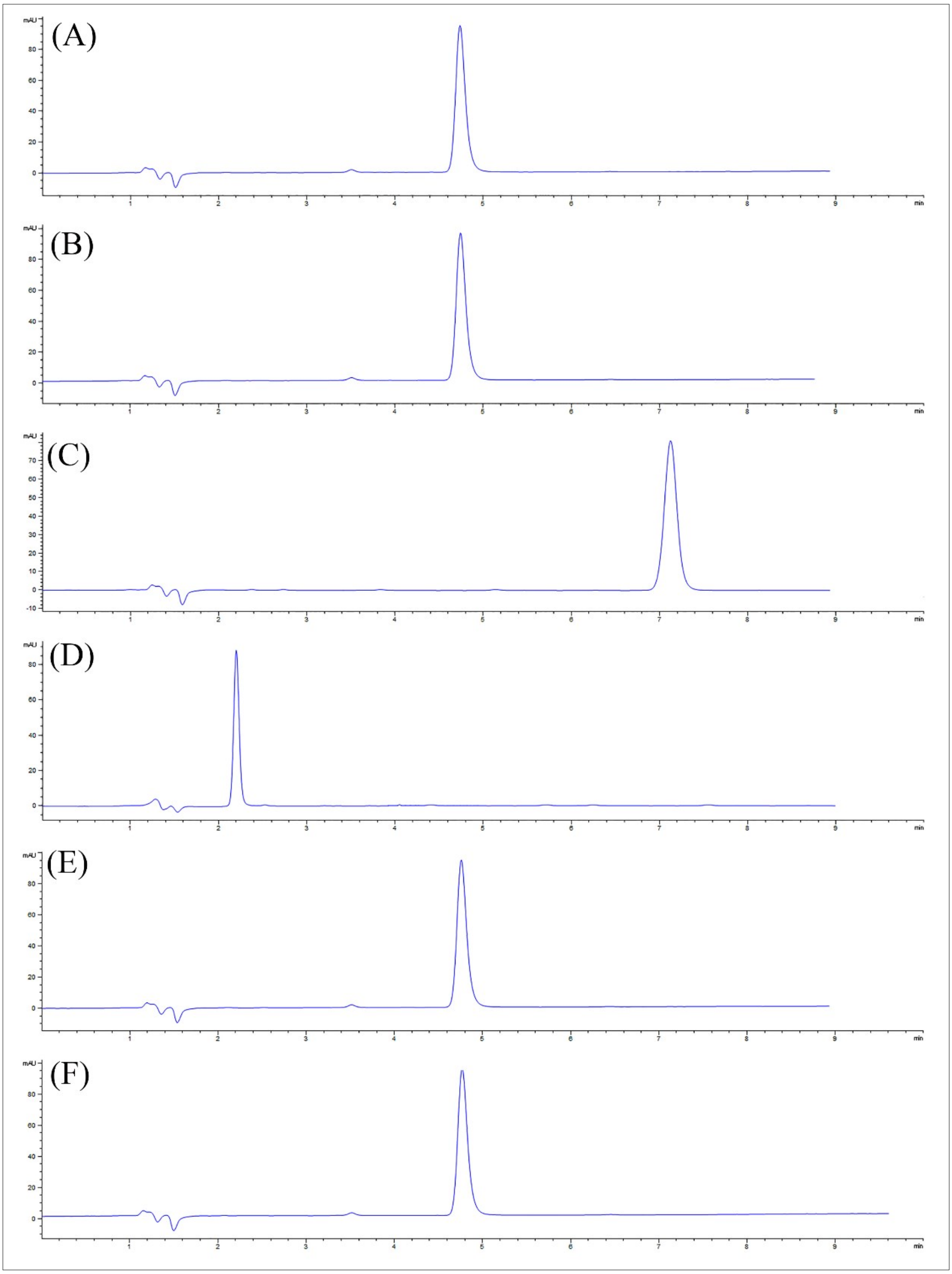


Fig. S4. HPLC chromatograms of the stability of solifenacin succinate [retention time (t_R) = 5.0 min] in different stress conditions, showing the disappearance of its peak after alkaline & oxidative degradation and the appearance of new peaks corresponding to SOL imp-I at t_R = 2.2 min and SOL imp-A at t_R = 7.6 min & where (A) intact drug; (B) acidic hydrolysis in 2 M HCl; (C) basic hydrolysis in 2 M NaOH; (D) oxidative degradation in 3% hydrogen peroxide; (E) photolytic degradation; (F) thermal degradation in 100°C oven.

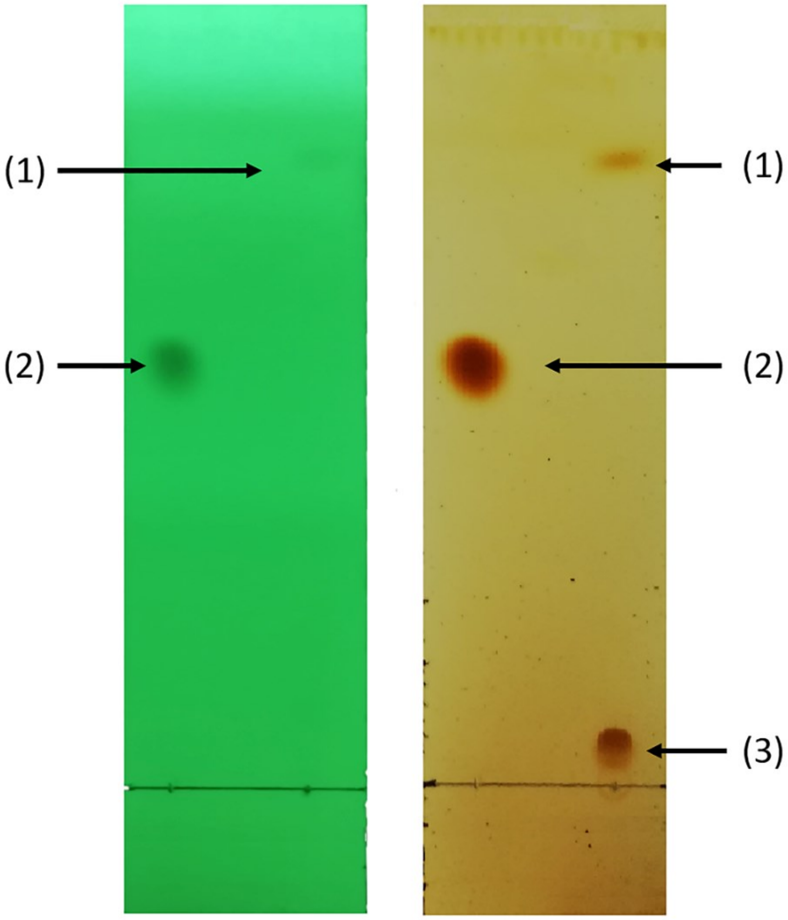


Fig. S5. HPTLC plate for SOL intact drug (2), along with its complete basic degradation products; SOL imp-A (1) under universal UV lamp (254 nm) and SOL imp-E (3) visualized only by iodine vapor.

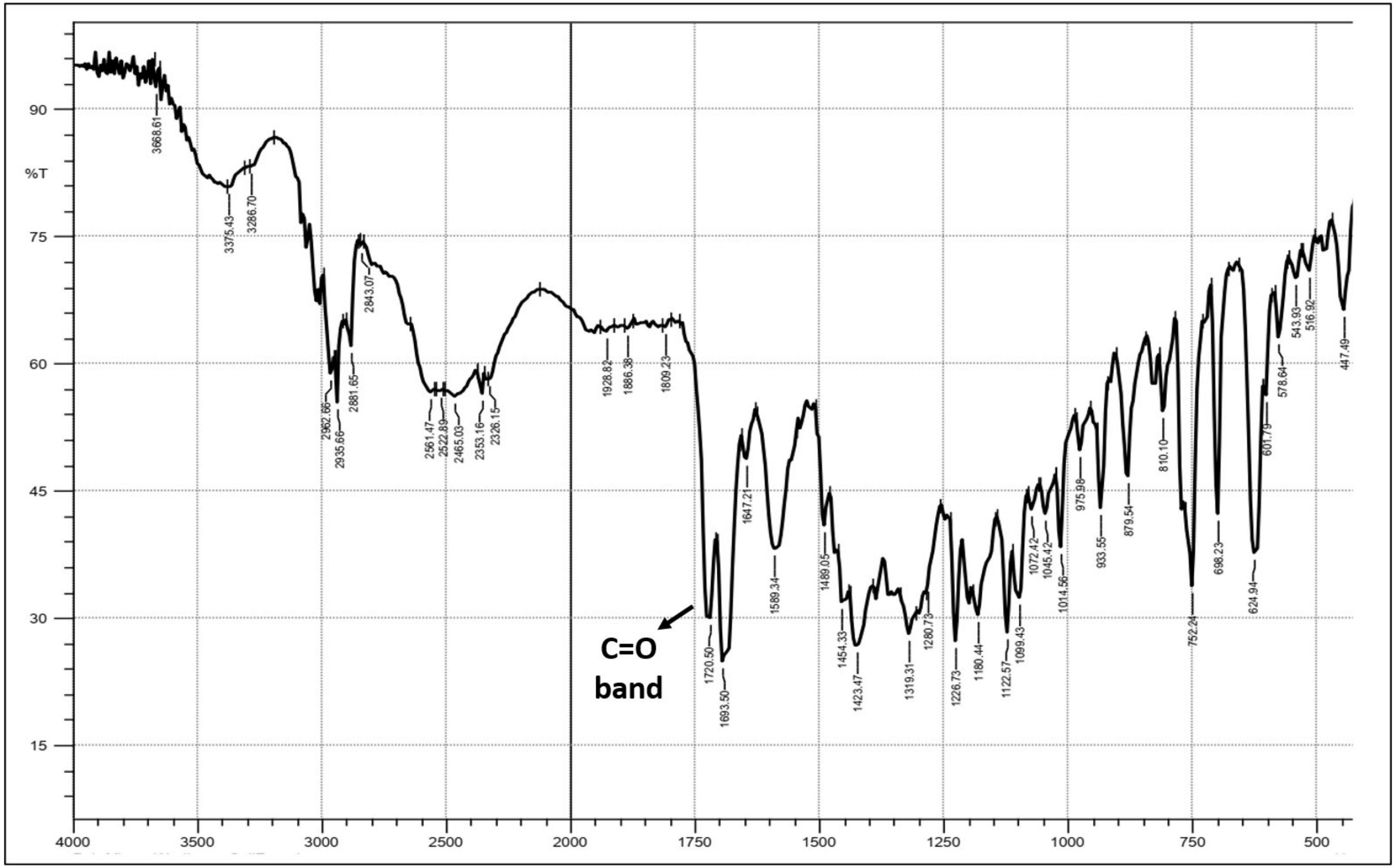


Fig. S6. (A) IR spectrum of SOL intact drug.

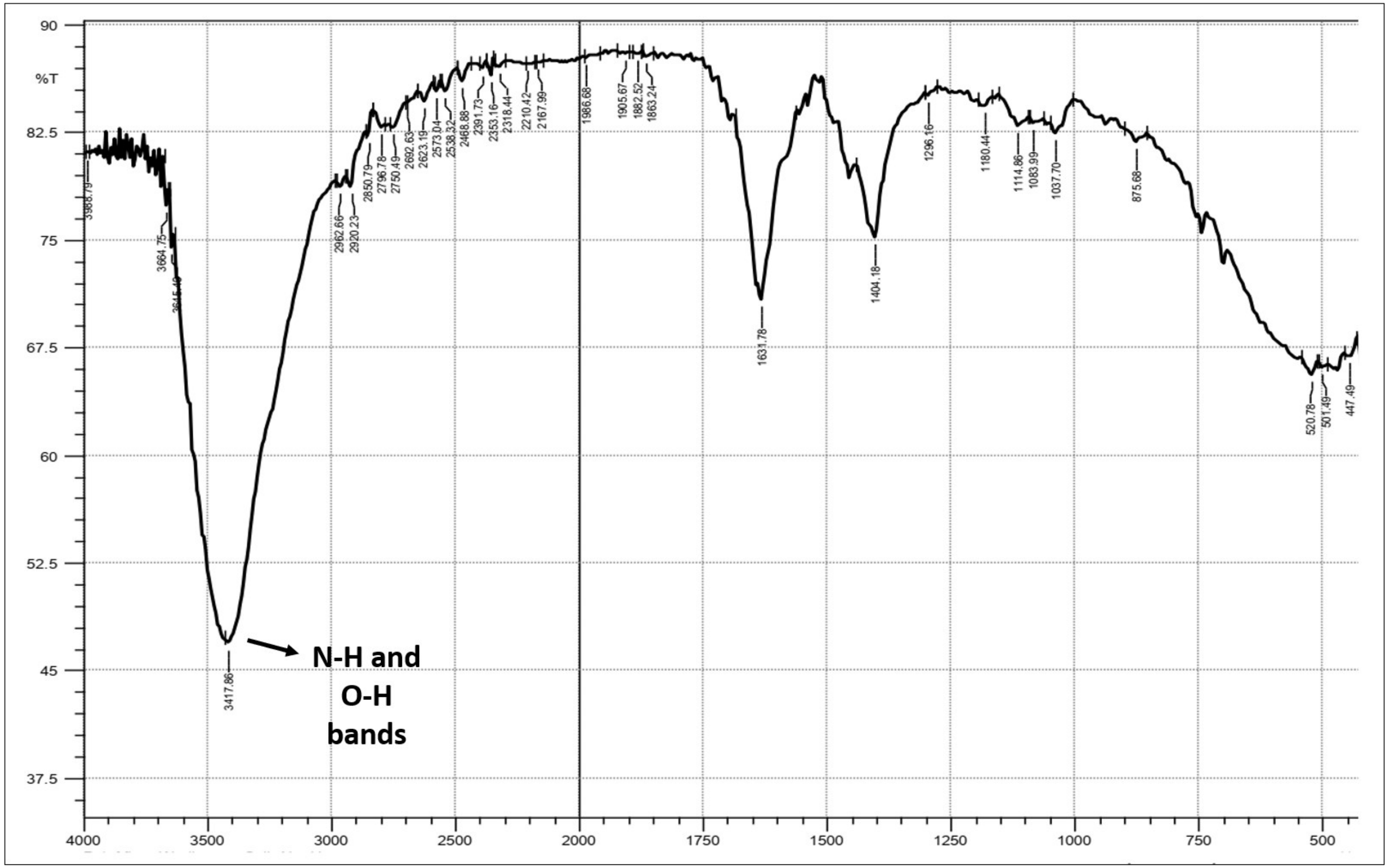


Fig. S6. (B) IR spectrum of SOL basic induced degradation products.

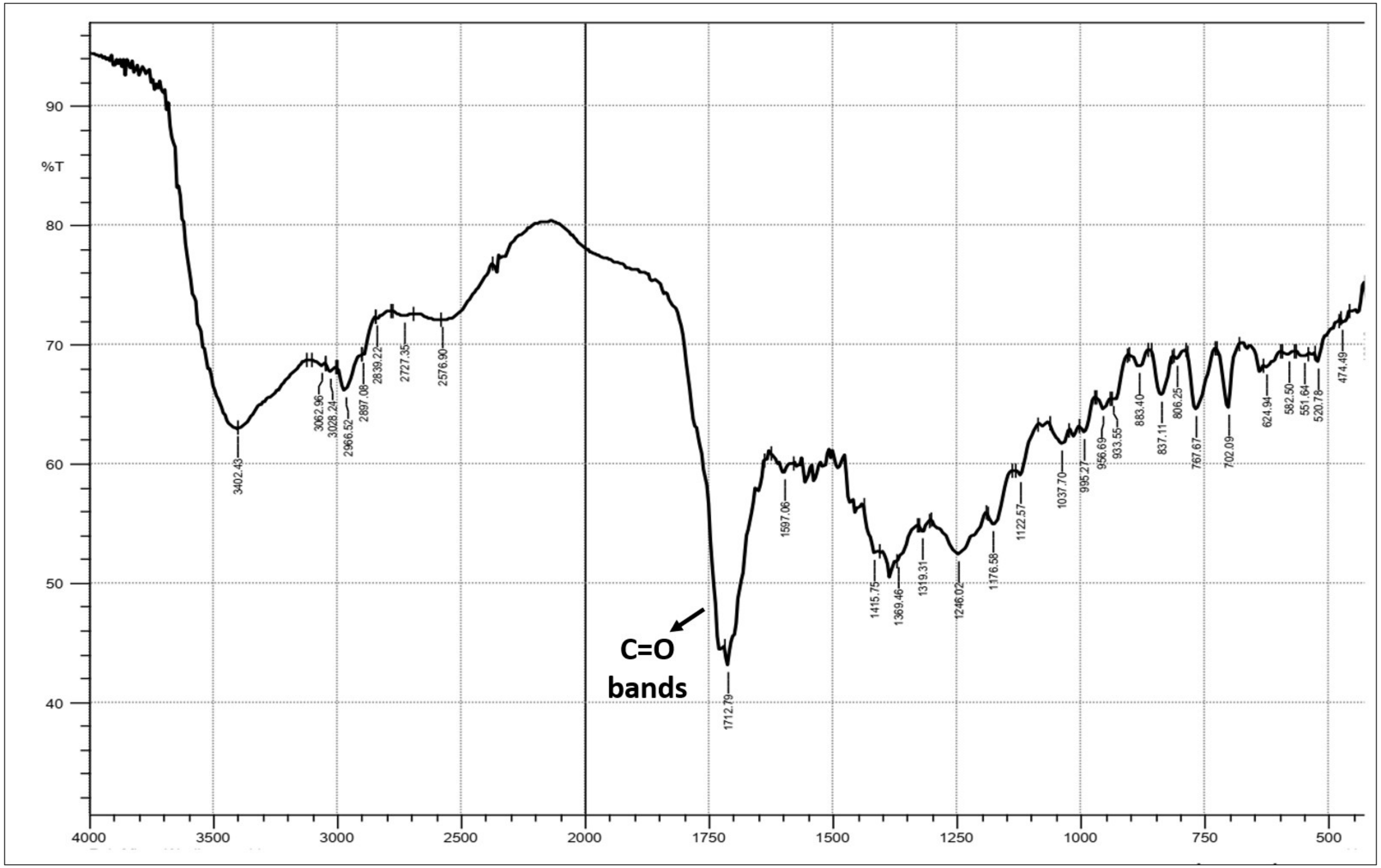


Fig. S6. (C) IR spectrum of SOL oxidative product.

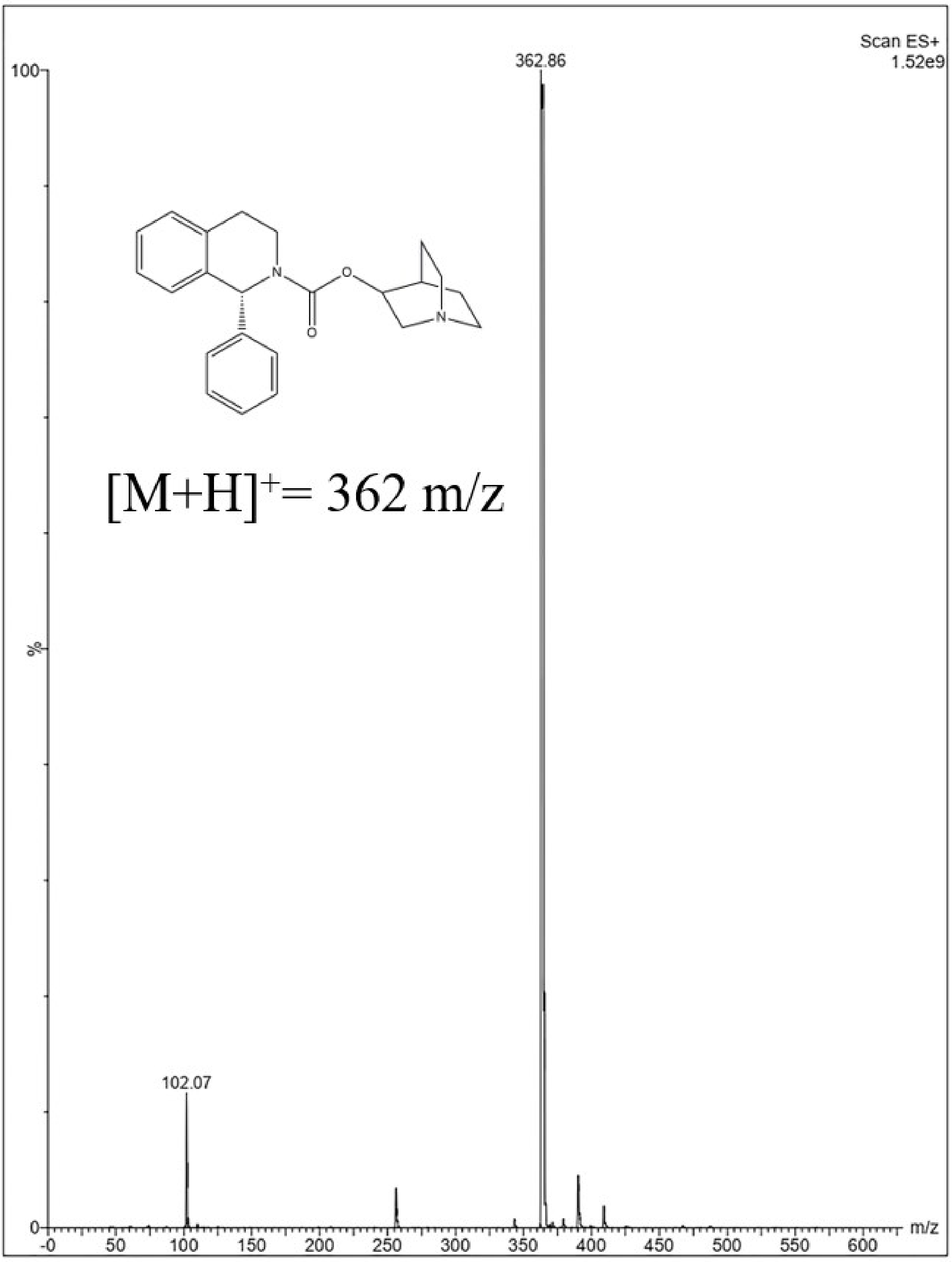


Fig. S7. (A) The mass spectrum showing molecular ion peak of SOL intact drug at 362 m/z.

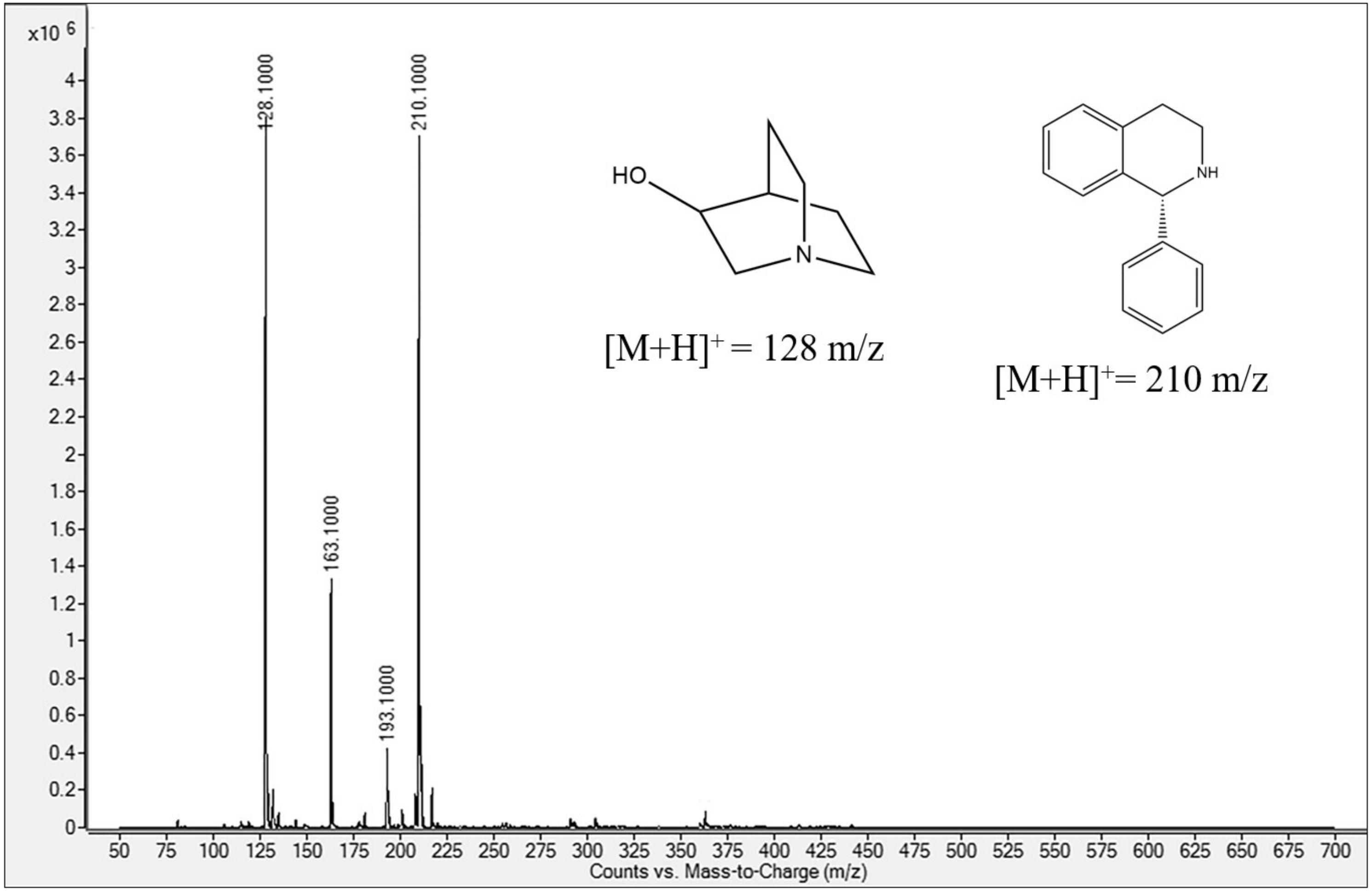


Fig. S7. (B) The mass spectrum showing molecular ion peak of two SOL basic degradation products; SOL imp-A & SOL imp-E at 210 m/z and 128 m/z, respectively.

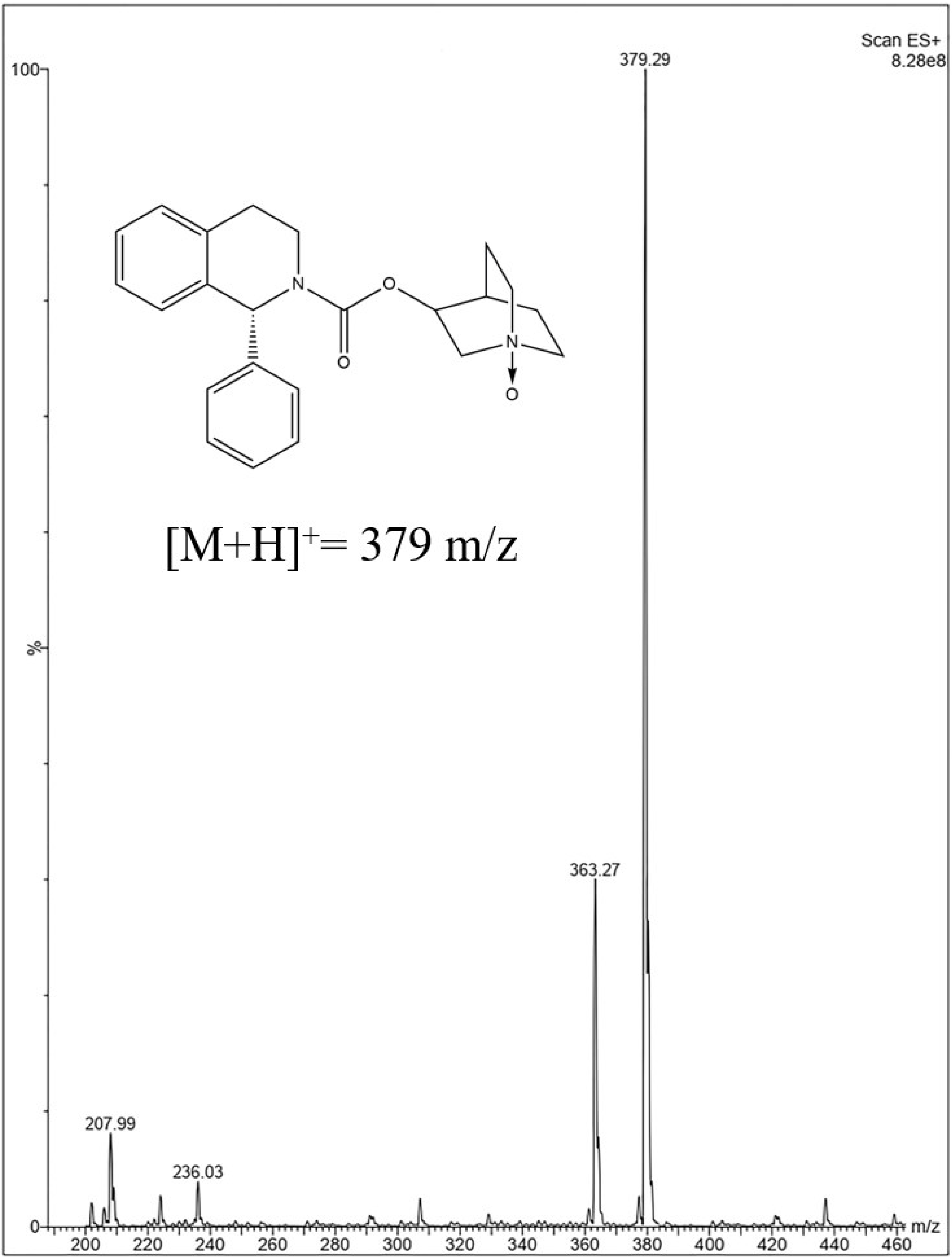


Fig. S7. (C) The mass spectrum showing molecular ion peak of SOL oxidative product; SOL imp-I at 379 m/z.

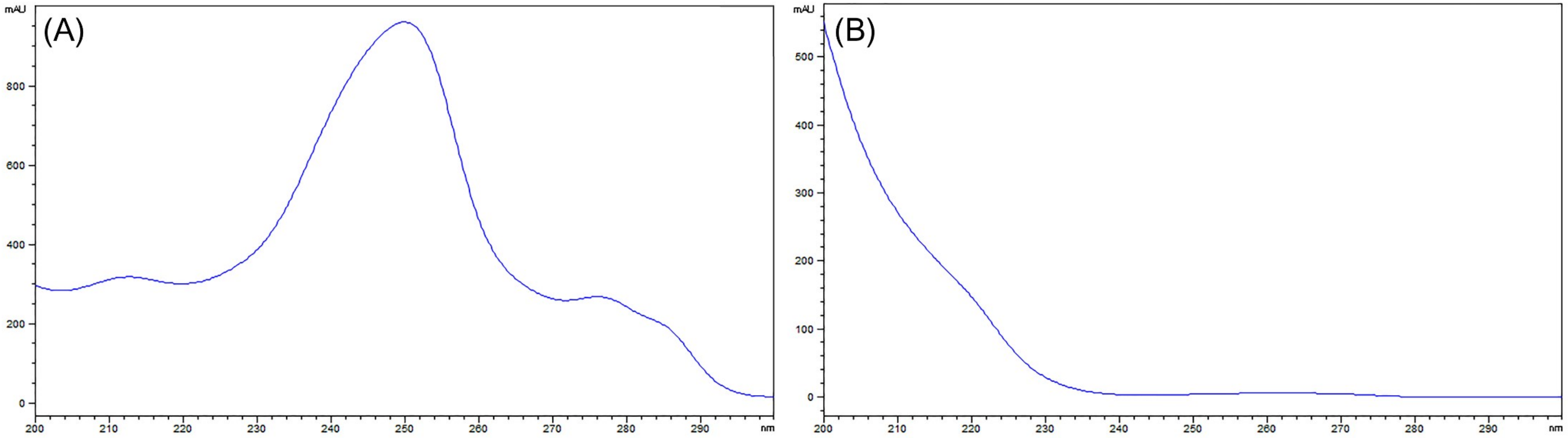
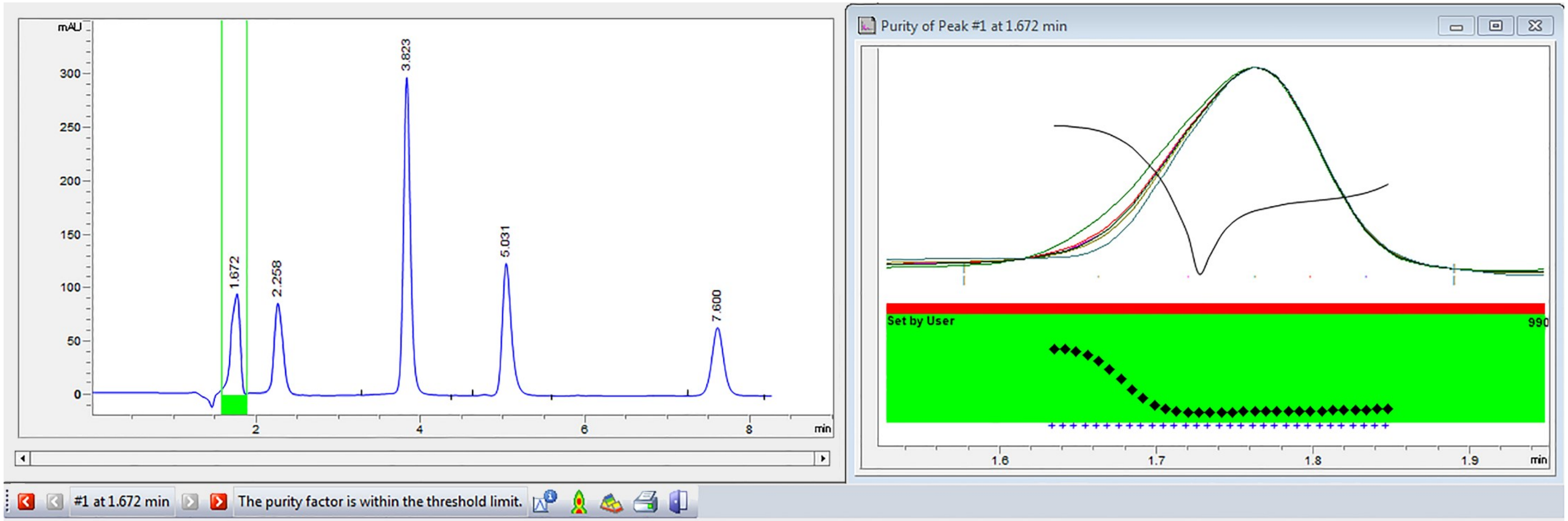


Fig. S8. On-line UV absorption spectra recorded by DAD detector for (a) alfuzosin HCl and (b) solifenacin succinate in the mobile phase.

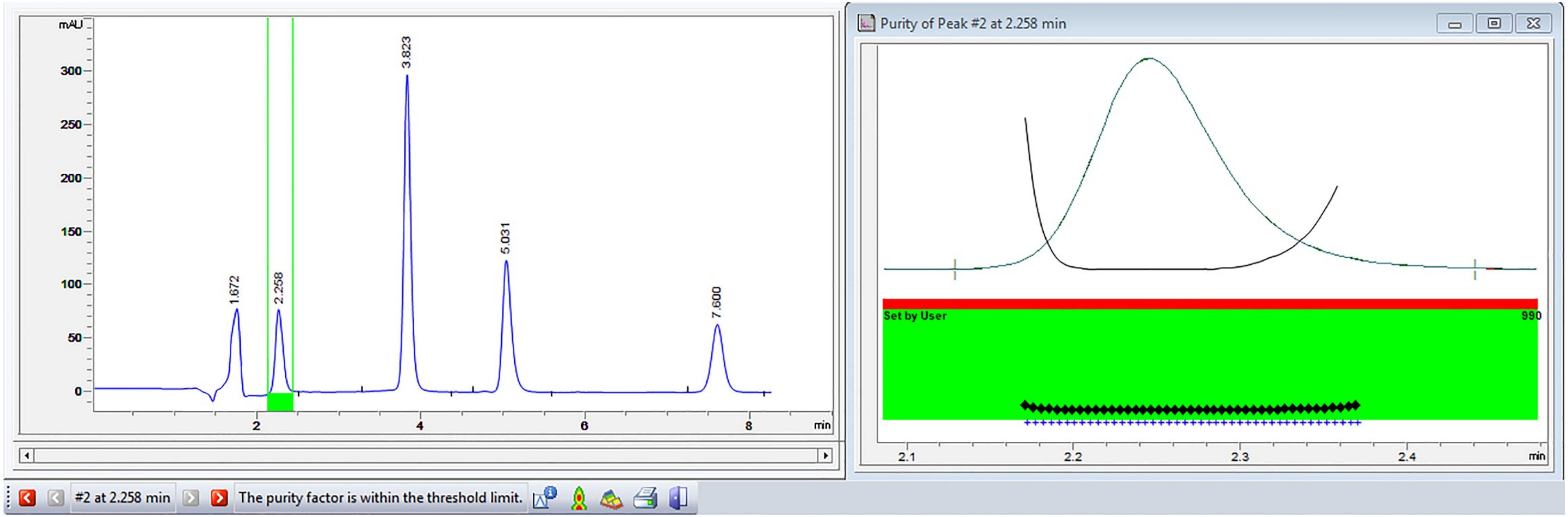


Purity of Peak #1 at 1.672 min

#1 at 1.672 min

The purity factor is within the threshold limit.

Fig. S9. (A) Peak purity assessment for ALF imp-D peak at t_R = 1.7 min showing purity factor of 995.822 at peak threshold 990.

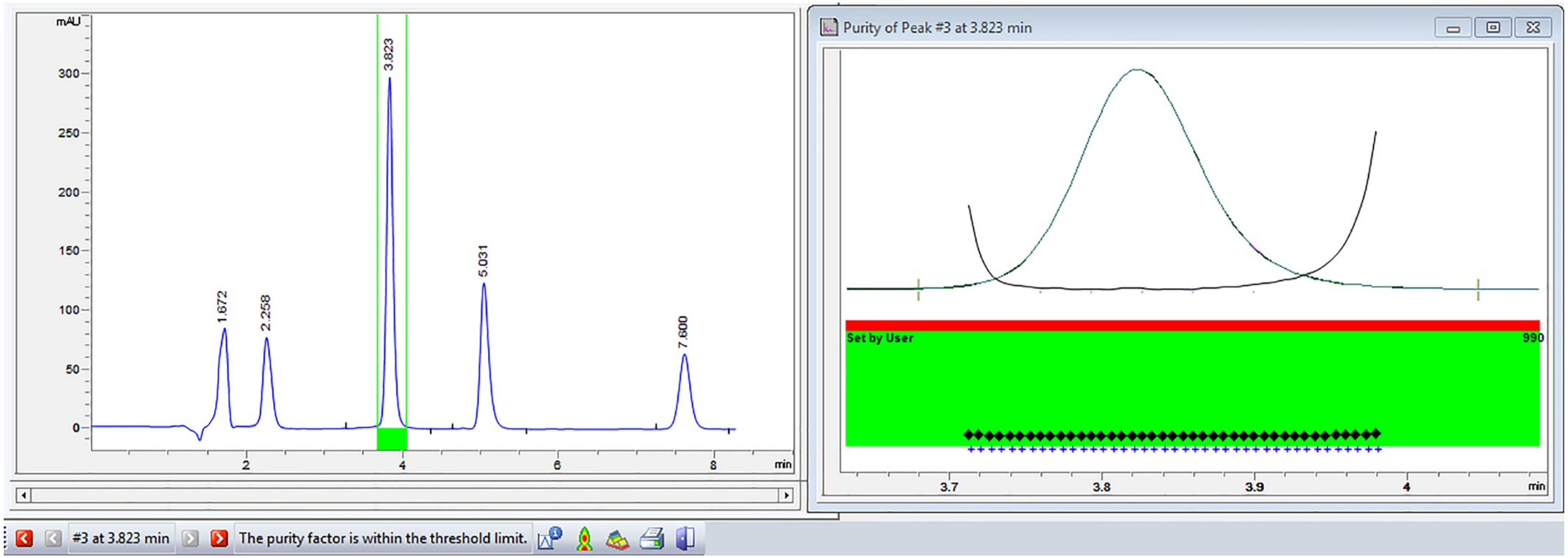


Purity of Peak #2 at 2.258 min

#2 at 2.258 min

The purity factor is within the threshold limit.

Fig. S9. (B) Peak purity assessment for SOL imp-I peak at t_R = 2.3 min showing purity factor of 999.912 at peak threshold 990.

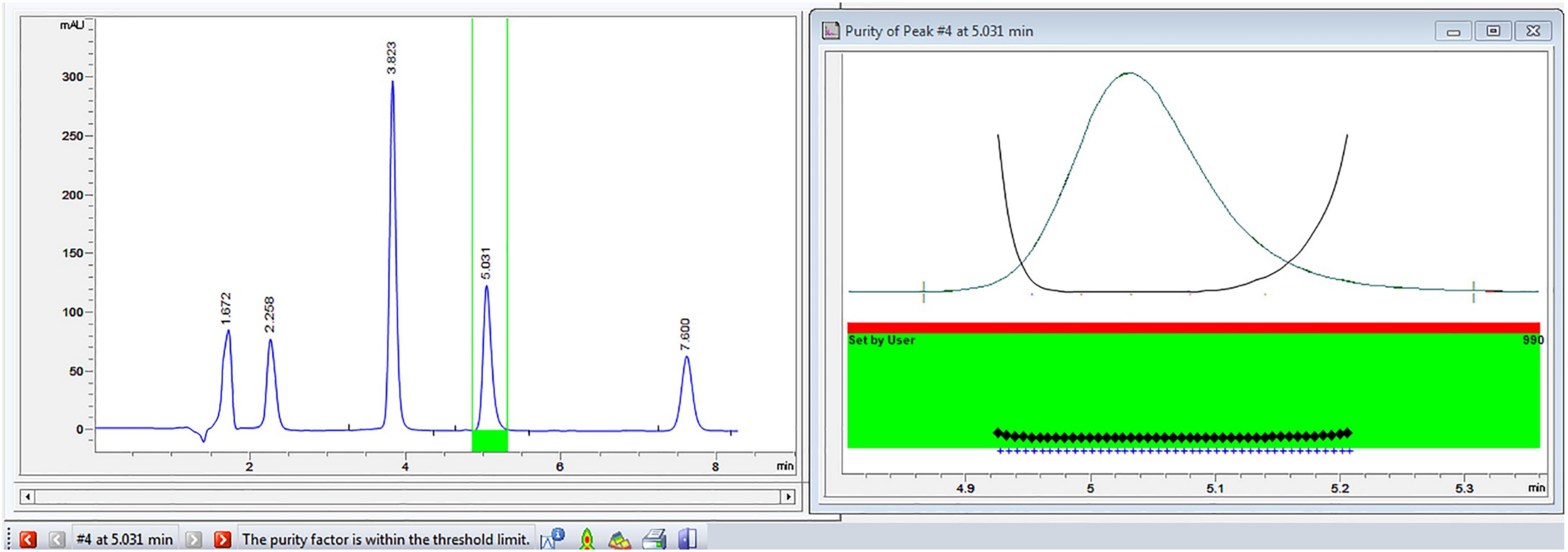


Purity of Peak #3 at 3.823 min

#3 at 3.823 min

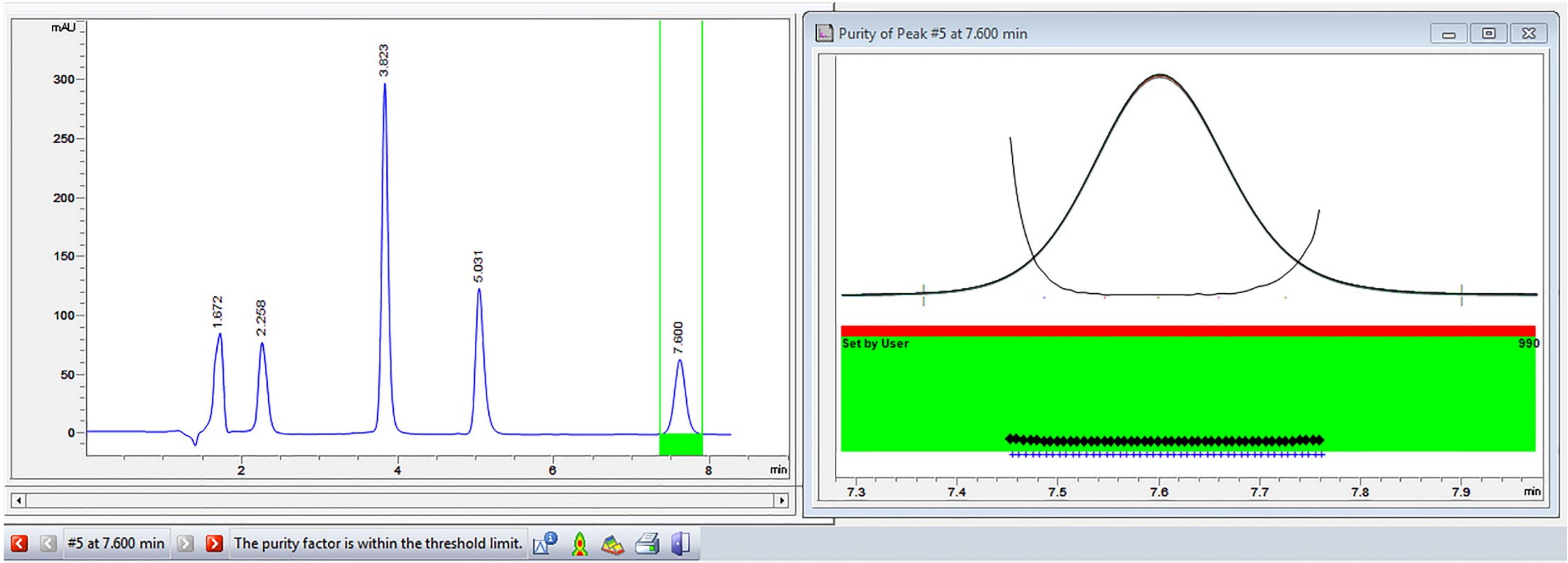
The purity factor is within the threshold limit.

Fig. S9. (C) Peak purity assessment for alfuzosin peak at t_R = 3.8 min showing purity factor of 999. 979 at peak threshold 990.



The purity factor is within the threshold limit.

Fig. S9. (D) Peak purity assessment for solifenacin peak at $t_R = 5.0$ min showing purity factor of 999.932 at peak threshold 990.



Purity of Peak #5 at 7.600 min

The purity factor is within the threshold limit.

Fig. S9. (E) Peak purity assessment for SOL imp-A peak at t_R = 7.6 min showing purity factor of 999.968 at peak threshold 990.

Table S1. Determination of ALF and SOL in its dosage form as well as accelerated stability-studied samples and application of standard addition technique using the proposed HPLC method.

Pharmaceutical formulation				ALF	SOL
Solitral® capsule (10mg ALF / 5mg SOL)		Found% [a] ± RSD%		100.47 ± 0.617	100.15 ± 1.573
Accelerated stability-studied samples				ALF	SOL
After 6 months		Found% [a] ± RSD%		99.27 ± 0.928	93.92 ± 1.907
Standard addition technique					
Taken (µg/mL)		Pure added (µg/mL)		Recovery %[b]	
ALF	SOL	ALF	SOL	ALF	SOL
30.0	15.0	15.0	7.5	99.17	98.42
30.0	15.0	30.0	15.0	99.86	99.29
30.0	15.0	60.0	30.0	101.93	99.37
		Mean ± RSD%		100.32 ± 1.236	99.03 ± 0.532

[a] Average of 5 determinations of capsule dosage form solution

[b] Average of three determinations

Table S2. Statistical comparison of the results obtained by the proposed HPLC method and official methods for the analysis of the two cited drugs.

Parameter	Proposed HPLC method		Official methods	
	ALF	SOL	ALF	SOL
Mean of recoveries	99.63	100.17	100.45	99.35
S.D.	0.839	1.003	0.410	1.695
Variance	0.704	1.006	0.168	2.873
n	5	5	5	5
Student's t-test (2.306) [a]	1.955	0.935	-------	-------
F-test (6.388) [a]	4.185	2.858	-------	-------

[a] The values in parentheses represent the corresponding tabulated values of t and F at p=0.05.