

## Supplementary Information

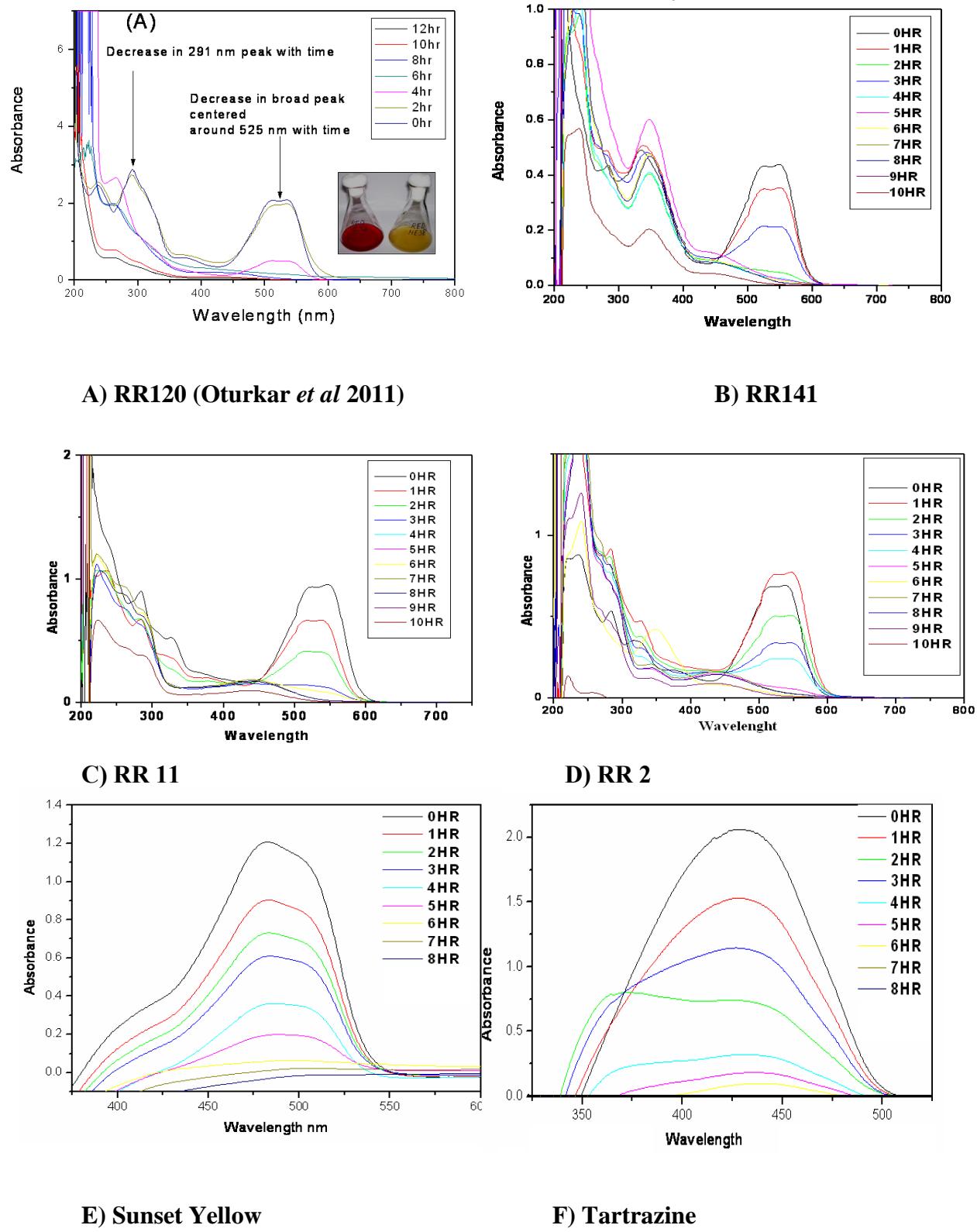
### **Synergistic action of flavin containing NADH dependant azoreductase and cytochrome P450 monooxygenase in azoaromatics mineralization**

**Chetan C. Oturkar<sup>a\*</sup>, Munif A. Othman<sup>b</sup>, Mahesh Kulkarni<sup>c</sup>, Datta Madamwar<sup>a</sup>,  
Kachru R. Gawai<sup>b\*\*</sup>**

<sup>a</sup>BRD School of Biosciences, Sardar Patel University, Vallabh Vidyanagar, India

<sup>b</sup>Department of Chemistry, University of Pune, India.

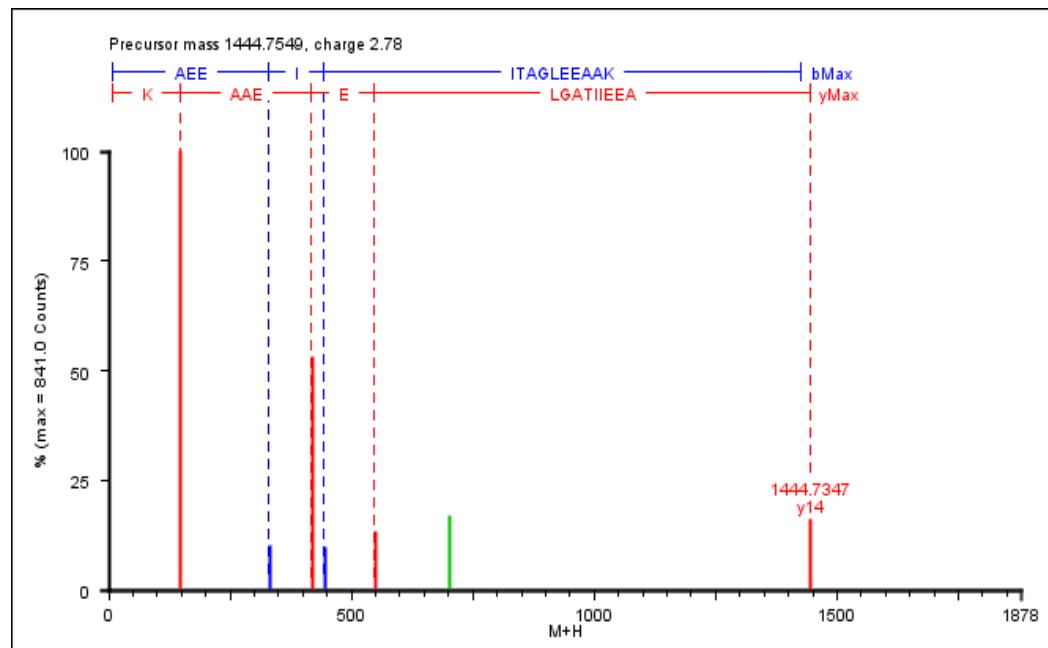
<sup>c</sup>Center for Material Characterization, National Chemical Laboratory Pune, India.



**Fig.S1** UV-Visible spectra during the decolorization azoic dyes

**Table S1 Substrate specificities study of purified azoreductase**

Substrates	Azoreductase Activity (U/mg)	$K_m$ ( $\mu\text{M}$ )	$V_{max}$ ( $\text{mmol} \cdot \text{min}^{-1} \cdot \text{mg}^{-1}$ )	$k_{cat}$ ( $\text{s}^{-1}$ )
RR120	$0.111 \pm 0.005$	$0.25 \pm 0.03$	$10.2 \pm 1.5$	$3.2 \pm 0.08$
RR11	$0.079 \pm 0.004$	$0.41 \pm 0.04$	$8.7 \pm 1.4$	$2.6 \pm 0.03$
RR5	$0.089 \pm 0.003$	$0.45 \pm 0.02$	$8.9 \pm 1.4$	$3.4 \pm 0.03$
RR141	$0.115 \pm 0.005$	$0.37 \pm 0.03$	$11.2 \pm 1.3$	$3.2 \pm 0.04$
SY	$0.105 \pm 0.004$	$0.38 \pm 0.02$	$9.3 \pm 1.7$	$4.1 \pm 0.02$

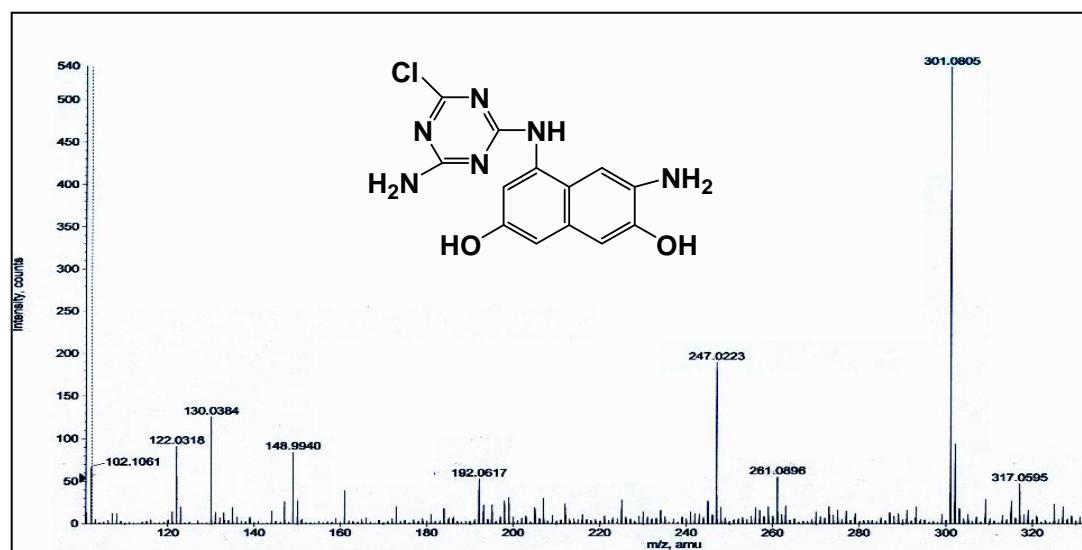


**Fig. S2 The fragmentation pattern of precursor  $\text{MH}^+$  (1444.753 Da) for peptide (K)AEEIIITAGLEEAAK(V)**

**Fig.S3 Possible major intermediates metabolites by GC-MS fragmentation spectra**

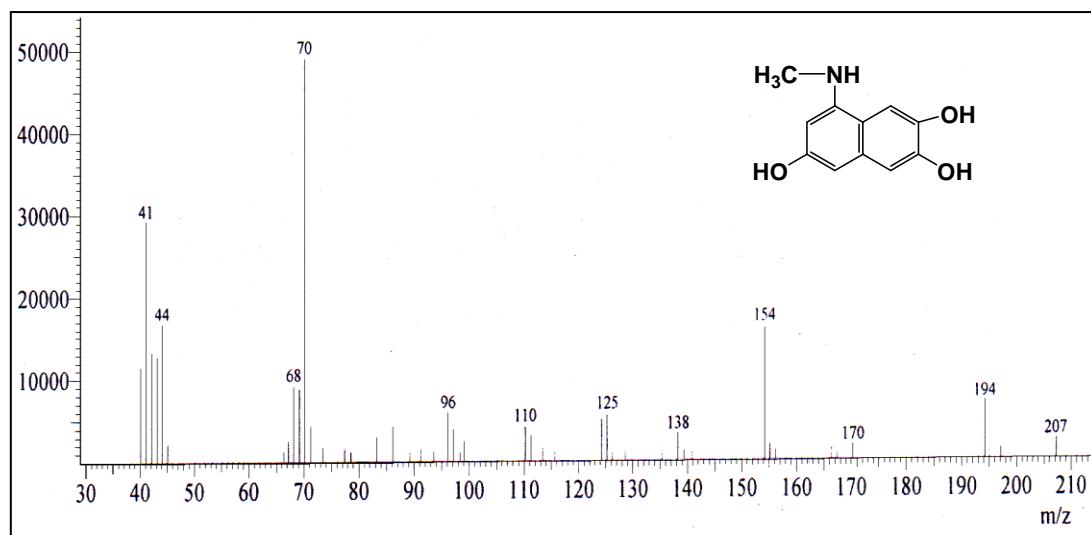
1) 5-(4-amino-6-chloro-1,3,5-triazin ylamino) 3-aminonaphthalene-2,7diol

GC-MS  $m/z$ : 317.05 [M $^+$ ], 301 [base peak]

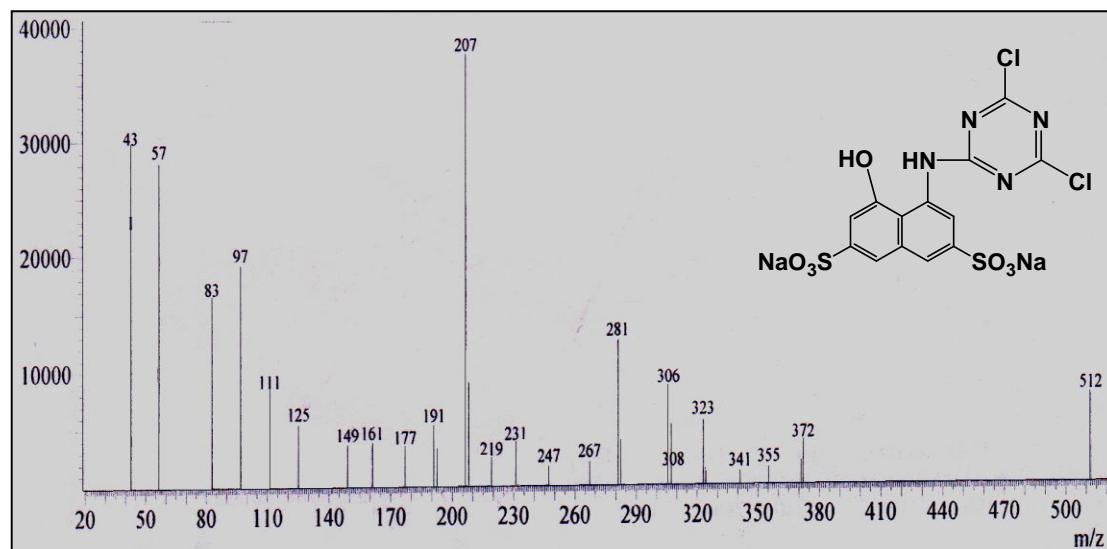


2) 8-(methylamino) naphthalene-2, 3, 6-triol

GC-MS  $m/z$ : 207.05 [M $^+$ ], 70 [base peak]

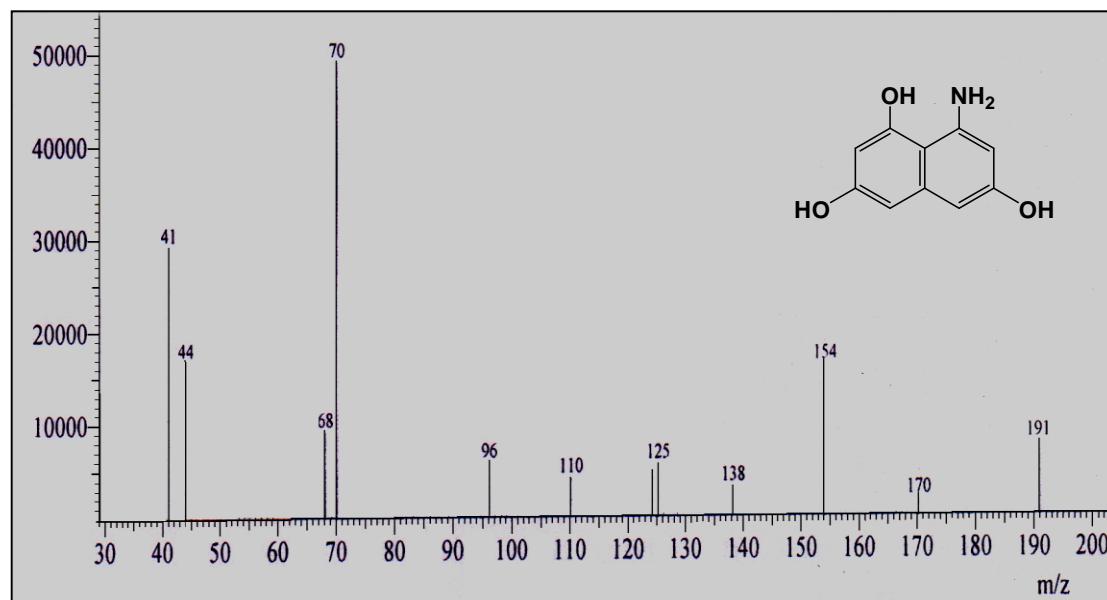


3) Sodium 4-(4,6-dichloro-1,3,5-triazin-2-ylamino)-5-hydroxynaphthalene-2,7-disulfonate  
GC-MS  $m/z$ : 512 [M $^+$ ], 207 [base peak]}



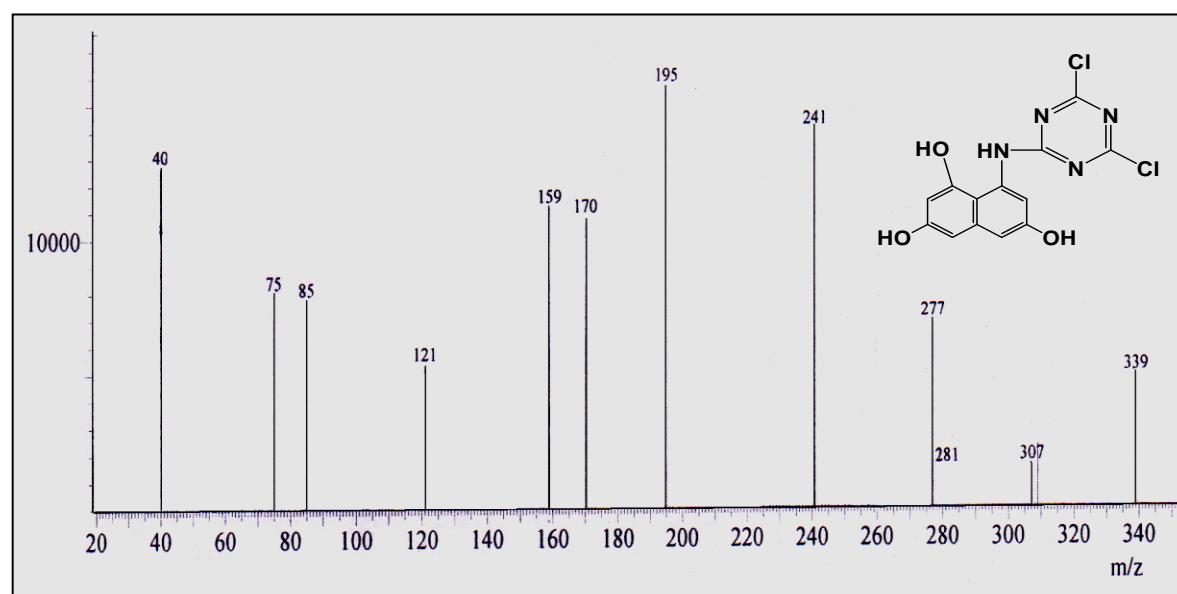
4) 8-aminonaphthalene-1, 3, 6-triol

GC-MS  $m/z$ : 191 [M $^+$ ], 70 [base peak]



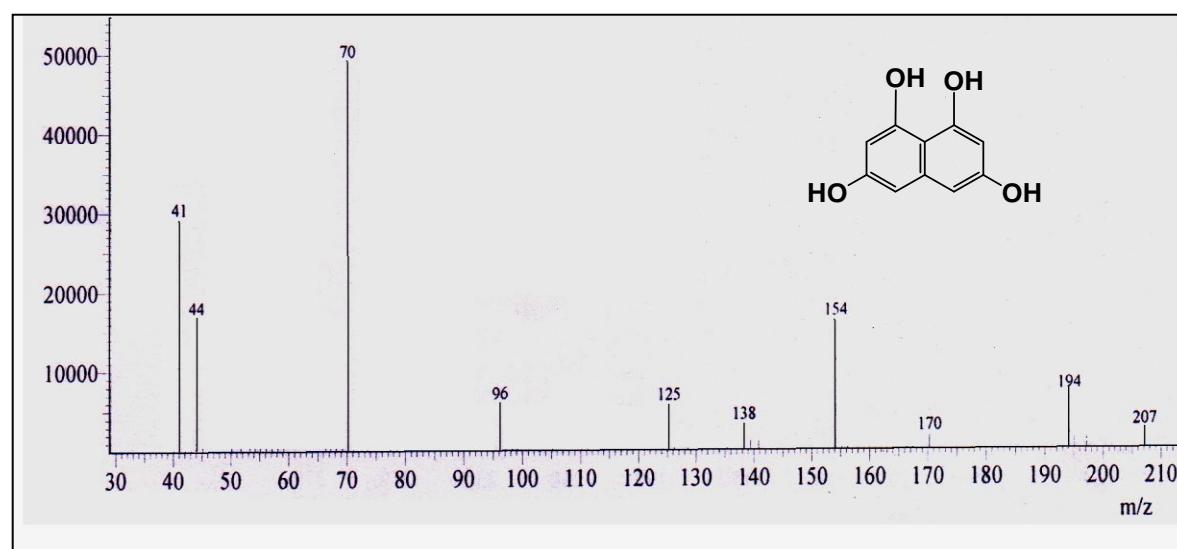
5) 8-(4,6-dichloro-1,3,5-triazin-2-ylamino) naphthalene-1,3,6-triol

GC-MS  $m/z$ : 339 [ $M^+$ ], 195 [base peak]



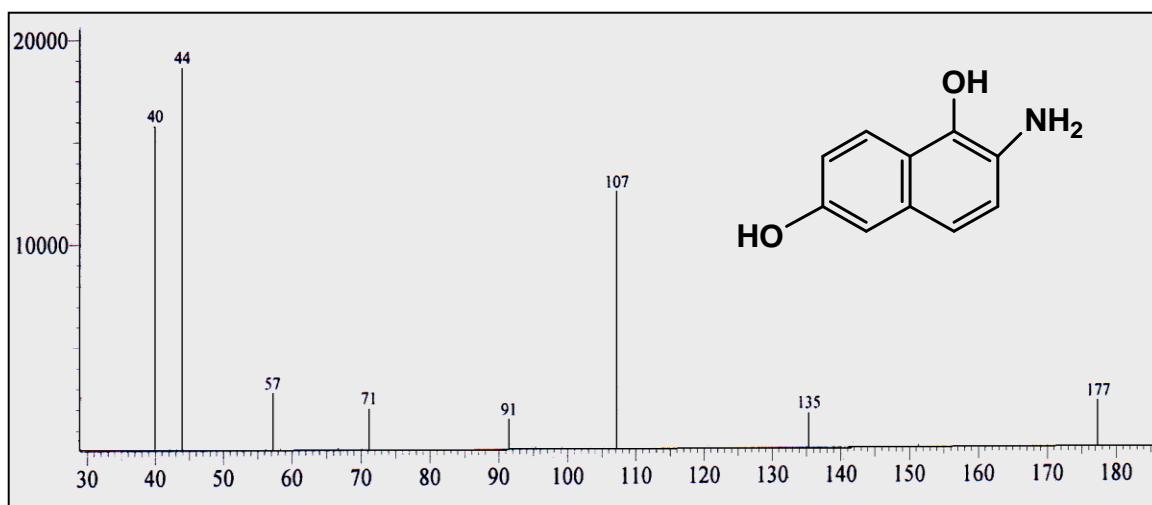
6) Naphthalene-1,3,6,8-tetraol

GC-MS  $m/z$ : 207 [ $M^+$ ], 70 [base peak]



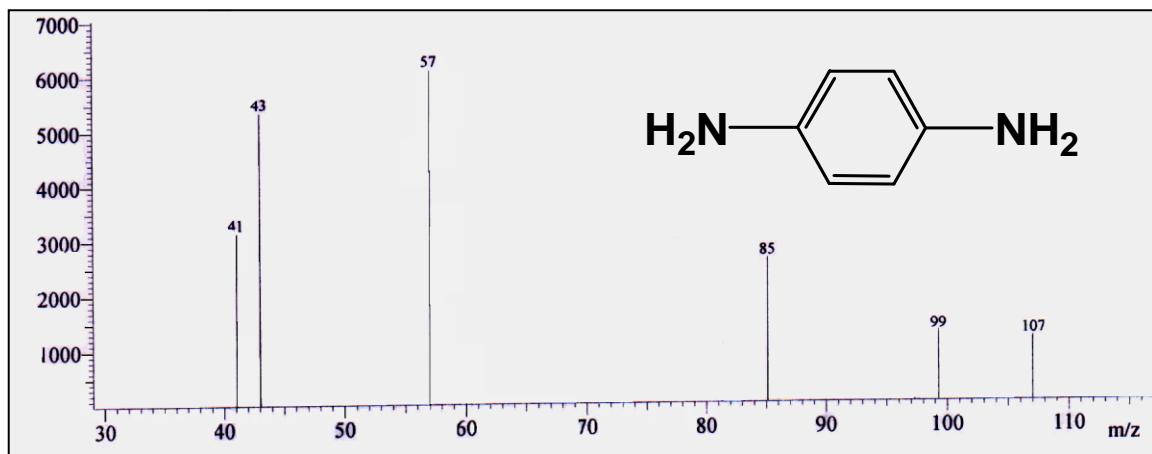
7) 2-aminonaphthalene-1,6diol

GC-MS  $m/z$ : 177 [ $M^+$ ], 44 [base peak]

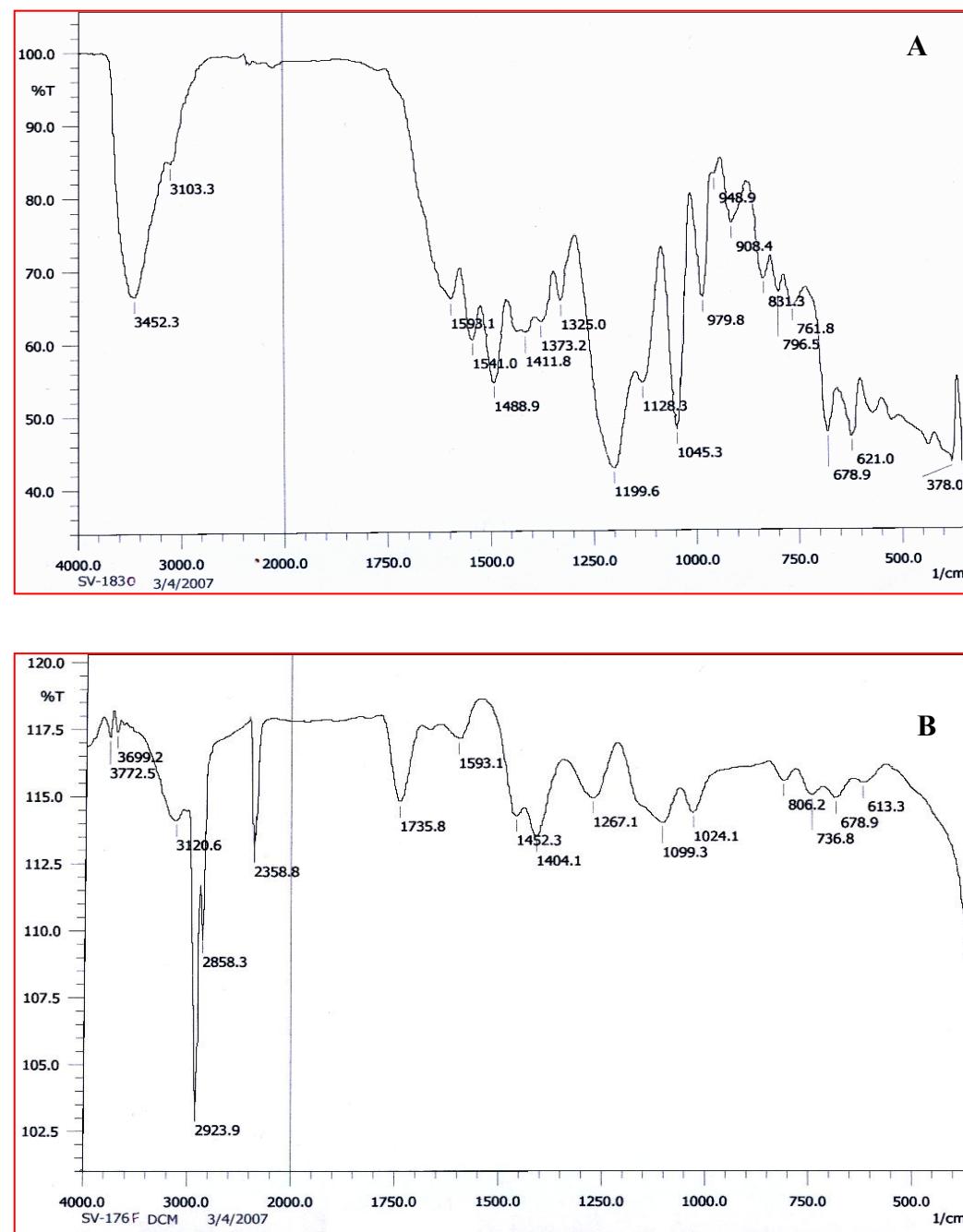


8) 1,4-diaminobenzene

GC-MS  $m/z$ : 107 [ $M^+$ ], 57 [base peak]

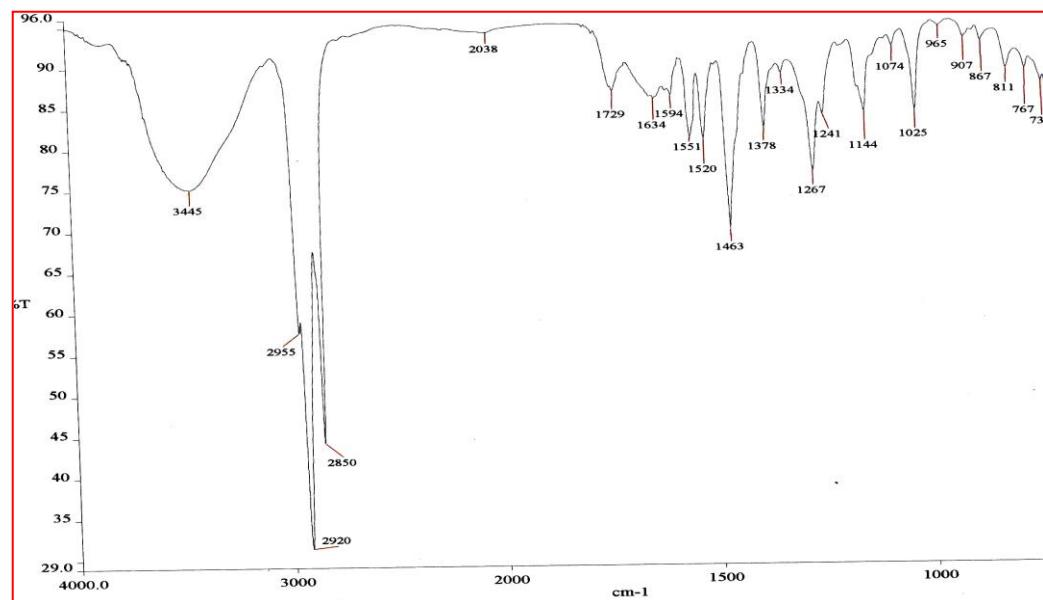


**Fig. S4 FTIR frequencies at A) 4 & B) 6 h of decolorisation/degradation**

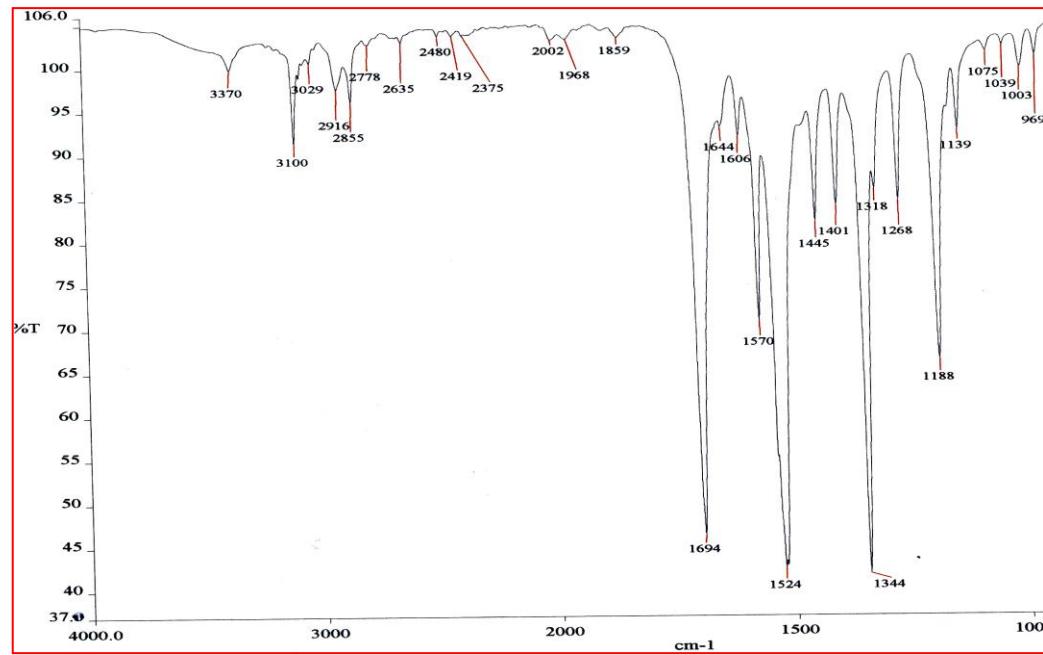


1) FTIR frequencies of RR141 A) 4 & B) 6 h of decolorisation/degradation

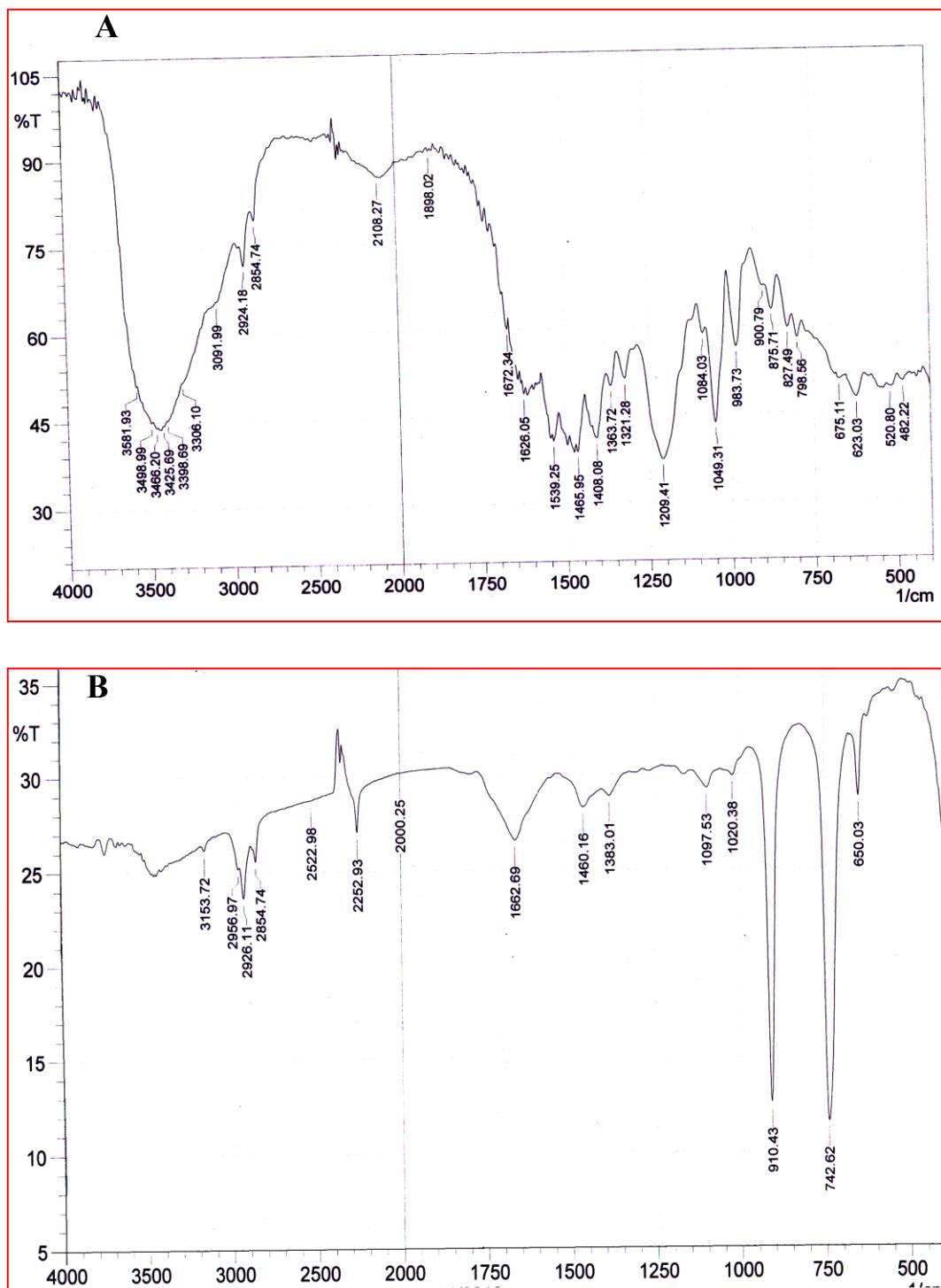
A



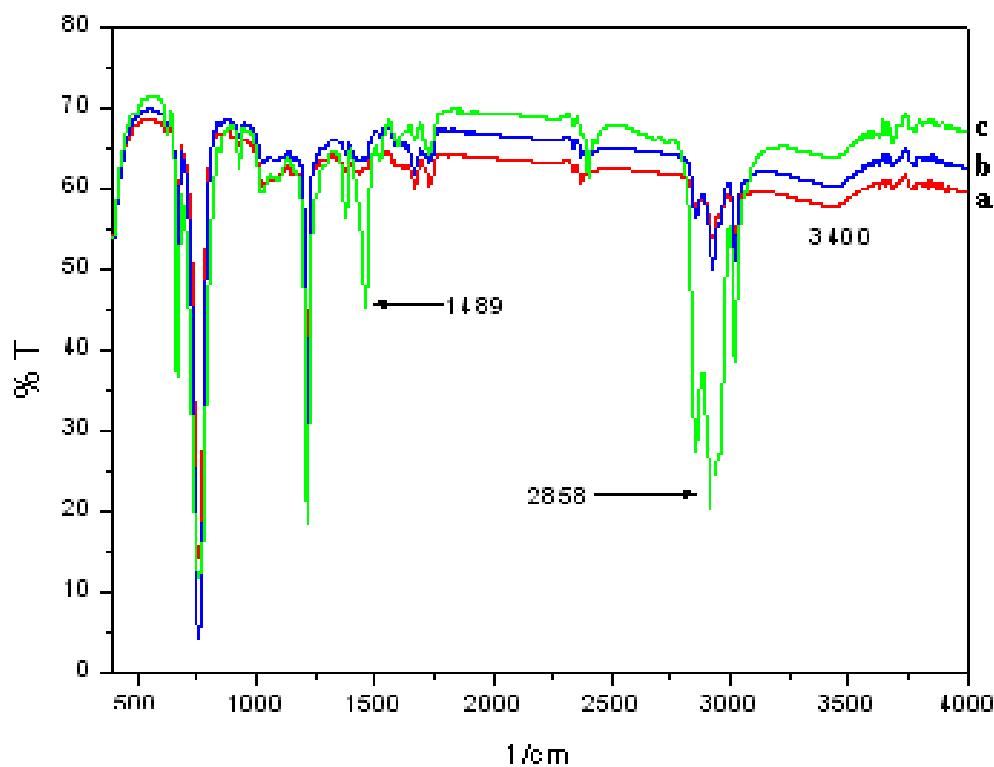
B



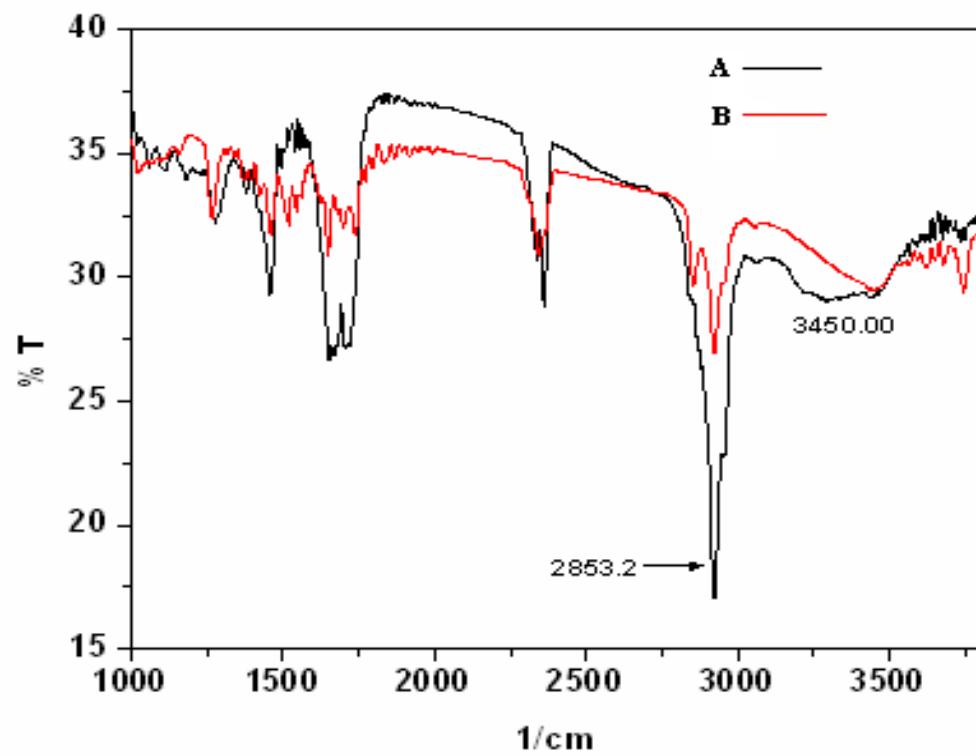
2) FTIR frequencies of RR 2 A) before degradation B) after degradation



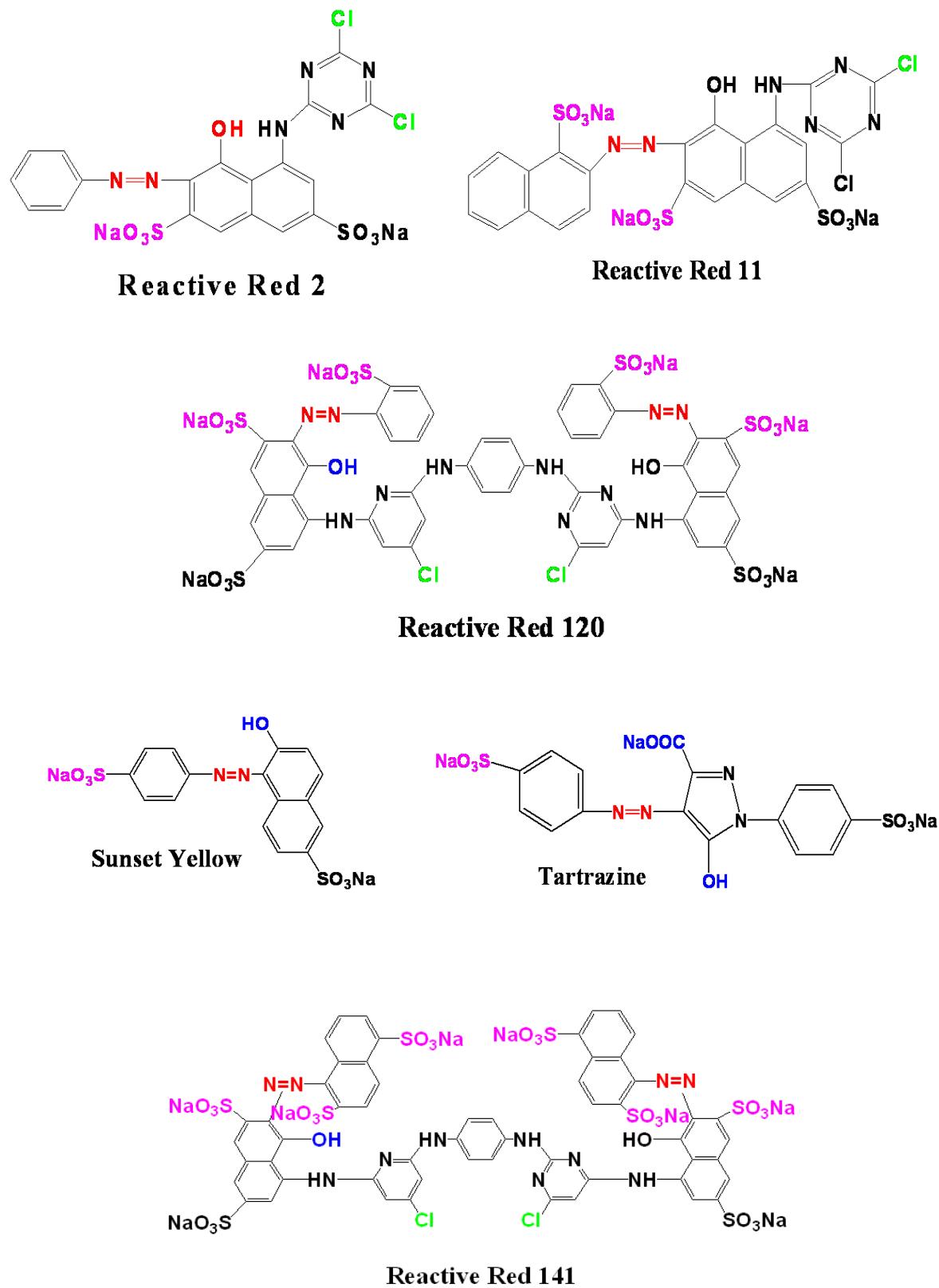
3) FTIR frequencies of RR 11 A) before degradation B) after degradation



4) FTIR frequencies of SY A) before degradation B) after degradation



5) FTIR frequencies of TAR A) before degradation B) after degradation



**Fig. S 5** Molecular structures of azo dyes