**C:\Users\Toshiba\Desktop\pectinase to be submitted\Figure 1AL.tif**

**Supplementary figure.1** Multiple sequence alignment using ClustalW. Sequences: EndopolygalacturonaseII from *Aspergillus niger* (PDB Id:1CZF), endopolygalacturonase G chain from *Colletotrichum lupini* (PDB Id:2IQ7), endopolygalacturonase I F chain from *Aspergillus niger* (PDB Id:1NHC), polygalacturonase from *Aspergillus aculeatus* (PDB Id:1IA5), endopolygalacturonase A chain from *Fusarium monoliforme* (PDB Id:1HG8), PecJKR01 from metagenome (GeneBank, NCBI accession number FJ538208). \* showing conserved cysteine residues. Square box showing site of mutations.

C:\Users\Toshiba\Desktop\pectinase to be submitted\Figure S1.tif

**Supplementary figure 2;** **(a)** 3D model structure of PecJKR01. Circle showing α helix structure placed above β-tunnel. Three parallel cysteine present in β-tunnel, out of which Cys-214 and Cys-230 formed possible disulphide linkage (S---S). **(b)** β -strands form tunnel like structure in 3D model structure of PecJKR01.

**Supplementary Table.1** Primers for introducing mutations in pecJKR01

|  |  |  |
| --- | --- | --- |
| **Primer Name** | **Primer Sequence** | **Purpose** |
| Rev-pecJKR01 | AAGTCGACTTAGGCTTTGTGTGAGTCATAG | for cloning in pQE 30; *Sal*1 site |
| Fwd-pecJKR01 | AAGGATCCATGAGTCTGCAGAAAATAAAAG | for cloning in pQE 30; *Bam*H1 site |
| **Primers to introduce cysteine** | | |
| F24C | CCTGATCGATCTTGCGATGTTACAGCG | for converting Phe24→Cys |
| REVF24C | CGCTGTAACATCGCAAGATCGATCAGG | for converting Phe24→Cys |
| A42C | GATGCGACGGGATGCATACAAAAGGCG | for converting Ala42→Cys |
| REVA42C | CGCCTTTTGTATGCATCCCGTCGCATC | for converting Ala42→Cys |
| M283C | GAACATAACGAATGCAAAGACGGGCAT | for converting Met283→Cys |
| REVM283C | ATGCCCGTCTTTGCATTCGTTATGTTC | for converting Met283→Cys |