# STRUCTURAL STUDIES OF LIPOXYGENASES FROM LEGUMES

*THESIS SUBMITTED TO THE UNIVERSITY OF MYSORE FOR THE DEGREE OF DOCTOR OF PHILOSOPHY IN BIOCHEMISTRY*

*BY*

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## DECLARATION

I hereby declare that the thesis entitled ***"Structural studies of Lipoxygenases from legumes"*** which is submitted herewith for the degree of ***Doctor of Philosophy*** in **Biochemistry** of the *University of Mysore*, *Mysore* is the result of the work done by me at the **Central Food Technological Research Institute, Mysore, India** in the **Department of Protein Chemistry and Technology** under the guidance of **Dr. A. G. Appu Rao** during the period July 2000 - March 2006.

I further declare the results of the work have not been previously submitted for any degree or fellowship.

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## CERTIFICATE

I hereby certify that this Ph.D. thesis entitled *"Structural studies of lipoxygenases from legumes"* submitted by Roopa Shree S. for the degree of *Doctor of Philosophy* in *Biochemistry* of the University of Mysore, Mysore, is the result of research work carried out by her in the *Department of Protein Chemistry and Technology, Central Food Technological Research Institute, Mysore, India,* under my guidance and supervision during the period 2000 – 2006. This has not been submitted either partially or fully to any degree or fellowship earlier.

**A.G. APPU RAO**
Guide

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## ABBREVIATIONS

| | |
|---|---|
| Ea | Energy of activation |
| ∆G* | Activational free energy change |
| ∆H* | Activational enthalpy change |
| ∆S* | Activational entropy change |
| ADH | Alcohol dehydrogenase |
| ANS | 8- Anilino 1-naphthalene sulphonic acid. |
| AOS | Allene oxide synthase |
| BCIP | 5-bromo-4-chloro-3-indolyl-phosphate |
| BSTFA | bis(trimethylsilyl) trifluoroacetamide |
| CA | Carbonic anhydrase |
| CAPS | 3-[cyclohexyl amine]-1-propane sulfonic acid |
| Da | Daltons |
| *D.biflorus* | *Dolichos biflorus.* |
| DBL | *Dolichos biflorus* lectin |
| DBSL | *Dolichos biflorus* seed lectin |
| DEA | Dihydroxyeicosatetraenoic acid |
| DES | Divinylether synthase |
| DOC | Deoxycholic acid. |
| EAS | Epoxy alcohol synthase |
| ETYA | 5,8,11,14 Eicosatetraynoic acid |
| FA-OOH | Fatty acid hydroperoxide |
| GalNAc | N-acetyl galactosamine |
| GC MS | Gas chromatography mass spectrometry. |
| GST | Glutathione S transferase |
| H(P)ETE | Hydro(pero)xyeicosatetraenoic acid |
| H(P)OD | Hydro(per)oxide |
| HGLOX | Horsegram lipoxygenase |
| HPL | Hydroperoxide lyase. |
| HU | Haemagglutination unit. |

| | |
|---|---|
| $IC_{50}$ | Mid point inhibitor concentration |
| JA | Jasmonic acid |
| Kcal | Kilo calories |
| kDa | kilo daltons |
| Ki | Inhibition constant |
| $K_m$ | Michaelis Menton Constant |
| $K_r$ | Rate constant. |
| LA | Linoleic acid |
| LDL | Low density lipoprotein |
| LOX | Lipoxygenase |
| LT | Leukotriene |
| MALDI-TOF/ MS | Matrix Assisted Laser Desorption ionization-Time of Flight/Mass Spectroscopy |
| NDGA | Nor-dihydorguaiaretic acid |
| OPDA | Oxo-phytodienoic acid. |
| PA | Periodic acid |
| PAGE | Poly acrylamide gel electrophoresis. |
| PC | Phosphatidyl Choline |
| PDB | Protein data bank |
| PG | Prostaglandin |
| POX | Peroxygenase. |
| PTH | Phenyl thio hydantoin |
| PUFA | Poly unsaturated fatty acid |
| PVDF | Polyvinyldiflouride. |
| RP-HPLC | Reverse phase high performance liquid chromatography |
| S | Substrate |
| SDS | Sodium dodecyl sulphate |
| SP-HPLC | Straight phase high performance liquid chromatography. |
| $T_m$ | Mid point temperature |
| TNS | 2,6,Toluidinylnaphthalenesulfonate |
| TPCK | L-1-Tosyl-amido-2-phenylethyl chloromethyl ketone |

| | |
|---|---|
| V | Velocity of the reaction |
| VOC | Volatile organic chemicals |

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