Supporting Information

Characterization of Differential Dynamics, Specificity, and Allostery of Lipoxygenase Family Members

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Supplementary Figures

Figure S1: Matrices and dendrograms showing the sequence (a) and structural (b) differences between the 88 LOXs in the PDB. (a) The pairwise fractional sequence identity (SID) is shown in the matrix (top) and the Hamming distance (1 – SID) is used for the sequence dendrogram (bottom). (b) The pairwise root mean-square deviation (RMSD) in Å is shown in the matrix and used for the dendrogram. Note that human LX15B (also known as 15LO2) is clustered together with human LOX5 in the sequence-based dendrogram (left, shown by the arrow), while it is closer to rabbit and pig LOX15 (or 15L01) in terms of its structure (arrow in right dendrogram).
Figure S2: Sequence alignment of human LOX12, LOX15 (15LO1), LX15B (15LO2), and *Pseudomonas aeruginosa* LOX (also known as LOXA or pLoxA). Residues conserved in at least three of the four sequences are colored. Hydrophobic residues are blue, polar residues are green, positively charged residues are red, negatively charged residues are purple, glycines are orange, and prolines are yellow. We used the Jalview\(^1\) program for visualization.
**Figure S3: Additional details about generic cross-correlations.** The left panel is a reproduction of Figure 3a with the column corresponding to the WxxAK motif highlighted. The right panel shows the values along this row in the context of the 3D structure. Residues in *dark red* show highly correlated (same direction) motions with this motif while regions in *dark blue* show highly anti-correlated (opposite direction) motions. The *red arrow* indicates L383, which corresponds to L367 in 12/15 Lipoxygenase that has been proposed to be near an oxygen access pathway.²
Figure S4: PRS results for 15LO1 alone (a) and in complex with PEBP1 (b). The top panels show the PRS matrices with the bars along the right and top axes representing the effectiveness and sensitivity, respectively, as in Figure 4. The middle panels show structures colored by sensitivity with the sensors in red and the bottom panels are colored by effectiveness with the effectors in red. PEBP1 is shown schematically in panel b for clarity. For a detailed structure of PEBP1-15LO1 complex see previous work\textsuperscript{3}. 
Figure S5: Generic mechanical stiffness properties from 88 LOX structures. (a) The mechanical stiffness (MechStiff) matrix shows the effective spring constant associated with uniaxial tension applied to each pair of residues (along the two axes). The entries are colored from red (stiffest) to blue (most flexible). The black bars at the top show the averages over each column, thus providing a residue-based metric for the resistance to deformation. (b) Ribbon diagram of LOX core domain (salmon). Regions distinguished by the strongest and weakest resistances to deformation are highlighted in red and blue, respectively, and indicated by the same color labels. Calculations were performed using the MechStiff module in ProDy\textsuperscript{4,5}. The semi-transparent portion displays the helical lid that is not present in mammalian LOXs.
## Supplementary Table

### Table S1: The 88 StructurallyResolved Lipoxigenases Used in This Study.

| PDB code | UniProtID | Name | # of aligned amino acids | SID [%] | RMSD [Å] |
|---------|-----------|------|--------------------------|---------|----------|
| 5medA   | B7JX99_CYAP8 | Arachidonate 15-LOX | 515 | 37.3 | 1.57 |
| 5megA   | B7JX99_CYAP8 | Arachidonate 15-LOX | 515 | 36.5 | 1.53 |
| 5mefA   | B7JX99_CYAP8 | Arachidonate 15-LOX | 514 | 36.9 | 1.59 |
| 5meeB   | B7JX99_CYAP8 | Arachidonate 15-LOX | 514 | 36.9 | 1.59 |
| 5medB   | B7JX99_CYAP8 | Arachidonate 15-LOX | 513 | 37.1 | 1.57 |
| 5meeA   | B7JX99_CYAP8 | Arachidonate 15-LOX | 515 | 37.1 | 1.60 |
| 5mefB   | B7JX99_CYAP8 | Arachidonate 15-LOX | 515 | 37.1 | 1.61 |
| 5megB   | B7JX99_CYAP8 | Arachidonate 15-LOX | 515 | 37.5 | 1.62 |
| 5ir4A   | LOXA_PSEAE  | Arachidonate 15-LOX | 518 | 99.6 | 0.49 |
| 5lc8A   | LOX_PSEAI   | Linoleate 9/13-LOX | 519 | 99.6 | 0.50 |
| 4g33A   | LOX_PSEAI   | Linoleate 9/13-LOX | 520 | 100.0 | 0.44 |
| 4rpeA   | LOX_PSEAI   | Linoleate 9/13-LOX | 519 | 99.8 | 0.43 |
| 4g32A   | LOX_PSEAI   | Linoleate 9/13-LOX | 520 | 100.0 | 0.11 |
| 5ir5A   | LOXA_PSEAE  | Arachidonate 15-LOX | 520 | 100.0 | 0.00 |
| 3fg4A   | AOSL_PLEHO  | Allene oxide synthase-LOX | 513 | 30.4 | 1.84 |
| 3fg1A   | AOSL_PLEHO  | Allene oxide synthase-LOX | 516 | 30.8 | 1.82 |
| 4qwtA   | AOSL_PLEHO  | Allene oxide synthase-LOX | 517 | 30.8 | 1.80 |
| 4qwtC   | AOSL_PLEHO  | Allene oxide synthase-LOX | 517 | 30.8 | 1.78 |
| 3fg1B   | AOSL_PLEHO  | Allene oxide synthase-LOX | 517 | 30.8 | 1.82 |
| 3fg3C   | AOSL_PLEHO  | Allene oxide synthase-LOX | 517 | 30.8 | 1.86 |
| 3fg3A   | AOSL_PLEHO  | Allene oxide synthase-LOX | 517 | 30.8 | 1.87 |
| 3fg3B   | AOSL_PLEHO  | Allene oxide synthase-LOX | 516 | 30.8 | 1.82 |
| 3fg3D   | AOSL_PLEHO  | Allene oxide synthase-LOX | 517 | 30.8 | 1.84 |
| 3fg1C   | AOSL_PLEHO  | Allene oxide synthase-LOX | 518 | 30.8 | 1.83 |
|   |   |   |   |   |   |
|---|---|---|---|---|---|
| 25 | 3fg4B | AOSL_PLEHO | Allene oxide synthase-LOX | 509 | 30.2 | 1.80 |
| 26 | 3fg4C | AOSL_PLEHO | Allene oxide synthase-LOX | 514 | 30.4 | 1.82 |
| 27 | 3fg4D | AOSL_PLEHO | Allene oxide synthase-LOX | 514 | 30.6 | 1.85 |
| 28 | 4qwtB | AOSL_PLEHO | Allene oxide synthase-LOX | 518 | 30.8 | 1.79 |
| 29 | 2fnqB | AOSL_PLEHO | Allene oxide synthase-LOX | 514 | 30.6 | 1.86 |
| 30 | 2fnqA | AOSL_PLEHO | Allene oxide synthase-LOX | 514 | 30.6 | 1.86 |
| 31 | 4nreA | LX15B_HUMAN | Arachidonate 15-LOX B | 515 | 30.2 | 1.85 |
| 32 | 3vf1B | Q2N410_9CNID | 11R-LOX | 510 | 27.9 | 2.11 |
| 33 | 3vf1A | Q2N410_9CNID | 11R-LOX | 510 | 27.9 | 2.11 |
| 34 | 2p0mA | LOX15_RABIT | Arachidonate 15-LOX | 505 | 24.8 | 2.09 |
| 35 | 1loxA | LOX15_RABIT | Arachidonate 15-LOX | 500 | 24.6 | 2.12 |
| 36 | 2p0mB | LOX15_RABIT | Arachidonate 15-LOX | 496 | 24.8 | 2.10 |
| 37 | 3rdeC | LOX15_PIG | Arachidonate 15-LOX | 508 | 25.4 | 1.88 |
| 38 | 3rdeA | LOX15_PIG | Arachidonate 15-LOX | 508 | 25.4 | 1.91 |
| 39 | 3rdeD | LOX15_PIG | Arachidonate 15-LOX | 509 | 25.4 | 1.89 |
| 40 | 3rdeB | LOX15_PIG | Arachidonate 15-LOX | 509 | 25.4 | 1.89 |
| 41 | 3d3lB | LOX12_HUMAN | Arachidonate 12-LOX, 12S-type | 414 | 22.9 | 2.29 |
| 42 | 3d3lA | LOX12_HUMAN | Arachidonate 12-LOX, 12S-type | 412 | 22.7 | 2.13 |
| 43 | 3v92B | LOX5_HUMAN | Arachidonate 5-LOX | 515 | 30.0 | 2.10 |
| 44 | 3v98A | LOX5_HUMAN | Arachidonate 5-LOX | 513 | 30.0 | 2.00 |
| 45 | 3v92A | LOX5_HUMAN | Arachidonate 5-LOX | 513 | 30.0 | 1.97 |
| 46 | 3o8yA | LOX5_HUMAN | Arachidonate 5-LOX | 514 | 30.0 | 2.02 |
| 47 | 3o8yB | LOX5_HUMAN | Arachidonate 5-LOX | 514 | 30.0 | 2.02 |
| 48 | 3v98B | LOX5_HUMAN | Arachidonate 5-LOX | 514 | 30.0 | 1.99 |
|   |   |   |   |   |   |   |
|---|---|---|---|---|---|---|
| 4 | 9 | 3v99A | LOX5_HUMAN | Arachidonate 5-LOX | 485 | 28.8 | 1.98 |
| 5 | 0 | 3v99B | LOX5_HUMAN | Arachidonate 5-LOX | 490 | 29.0 | 1.78 |
| 5 | 1 | 1jnqA | LOX3_SOYBN | Seed linoleate 9S-LOX-3 | 508 | 27.7 | 2.30 |
| 5 | 2 | 1hu9A | LOX3_SOYBN | Seed linoleate 9S-LOX-3 | 508 | 27.7 | 2.29 |
| 5 | 3 | 1no3A | LOX3_SOYBN | Seed linoleate 9S-LOX-3 | 506 | 27.9 | 2.24 |
| 5 | 4 | 1lnhA | LOX3_SOYBN | Seed linoleate 9S-LOX-3 | 505 | 27.5 | 2.28 |
| 5 | 5 | 1n8qA | LOX3_SOYBN | Seed linoleate 9S-LOX-3 | 508 | 27.9 | 2.26 |
| 5 | 6 | 1rrhA | LOX3_SOYBN | Seed linoleate 9S-LOX-3 | 507 | 28.3 | 2.20 |
| 5 | 7 | 1ik3A | LOX3_SOYBN | Seed linoleate 9S-LOX-3 | 504 | 28.1 | 2.20 |
| 5 | 8 | 1rrlA | LOX3_SOYBN | Seed linoleate 9S-LOX-3 | 508 | 28.5 | 2.30 |
| 5 | 9 | 1fgrA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 505 | 26.9 | 2.13 |
| 6 | 0 | 1fgoA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 505 | 27.1 | 2.13 |
| 6 | 1 | 1fgmA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 504 | 26.9 | 2.10 |
| 6 | 2 | 1ygeA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 507 | 27.1 | 2.24 |
| 6 | 3 | 2sblA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 505 | 27.3 | 2.12 |
| 6 | 4 | 2sblB | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 506 | 27.3 | 2.14 |
| 6 | 5 | 3bncA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 506 | 27.1 | 2.16 |
| 6 | 6 | 4whaA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 506 | 26.9 | 2.16 |
| 6 | 7 | 3bncA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 505 | 27.1 | 2.15 |
| 6 | 8 | 3pzwA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 507 | 26.9 | 2.27 |
| 6 | 9 | 1f8nA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 508 | 26.9 | 2.28 |
| 7 | 0 | 5t5vA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 507 | 27.1 | 2.17 |
| 7 | 1 | 3bndA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 508 | 26.9 | 2.20 |
| 7 | 2 | 1fgtA | LOX1_SOYBN | Seed linoleate 13S-LOX-1 | 504 | 26.9 | 2.07 |
|    | Identifier | Structure | Description | Size | RMSD | SID  |
|----|------------|-----------|-------------|------|------|------|
| 7  | 5seeA      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 504  | 27.3 | 2.12 |
| 7  | 5tr0A      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 495  | 26.7 | 2.11 |
| 7  | 1fgqA      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 505  | 27.5 | 2.11 |
| 7  | 4wfoA      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 505  | 27.3 | 2.10 |
| 7  | 3bneA      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 503  | 27.9 | 2.14 |
| 7  | 5t5vB      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 501  | 28.3 | 2.07 |
| 7  | 5tqpB      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 507  | 26.5 | 2.25 |
| 8  | 5tqoB      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 505  | 26.5 | 2.14 |
| 8  | 5tqoA      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 504  | 26.3 | 2.13 |
| 8  | 5tqpA      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 508  | 26.7 | 2.29 |
| 8  | 5tqnA      | LOX1_SOYBN| Seed linoleate 13S-LOX-1| 504  | 27.1 | 2.10 |
| 8  | 1rrlB      | LOX3_SOYBN| Seed linoleate 9S-LOX-3| 510  | 27.7 | 2.33 |
| 8  | 5fx8A      | F2QXM5_KOMPC| Manganese LOX| 500  | 21.0 | 3.34 |
| 8  | 5fnoB      | MNLOX_MAGO7| Manganese LOX| 503  | 21.2 | 3.22 |
| 8  | 5fnoA      | MNLOX_MAGO7| Manganese LOX| 503  | 21.5 | 3.20 |
| 8  | 5fx8B      | F2QXM5_KOMPC| Manganese LOX| 494  | 20.8 | 3.21 |

*a The first four characters are the identifier for the structure in the Protein Data Bank (PDB) and the fifth is the chain identifier.
*b corresponding record in UniProt. The part before the underscore is an identifier that is often an abbreviation for the protein and the part after the underscore corresponds to the organism from which it is derived.
*c Long names from UniProt.
*d number of aligned amino acid residues that form the conserved core whose structure and dynamics are compared using PCA and GNM analysis.
*e Percent sequence identity (SID) of core residues with respect to the pLOXA reference structure in row 14 (bold).
*f Root-mean-square deviation (RMSD) between core structure and the pLOXA reference structure in row 14 (bold).
Table S2. Key residues dominating the dynamics and allostery of LOX family members

| pLoxA                        | 15LO1  | 15LO2  | LOX12  |
|------------------------------|--------|--------|--------|
| H382                         | H365   | H378   | H365   |
| H377                         | H360   | H373   | H360   |
| H555                         | H540   | H553   | H540   |
| N559                         | H554   | S557   | N554   |
| I685                         | I662   | I676   | I663   |
| W357xxAK<sub>361</sub>      | W340-K344 | W353-K357 | W340-K344 |
| P228-R248                    | W222-R242 | W232-R252 | W222-R242 |
| P242-N243                    | A236-N237 | L246-N247 | A236-N237 |
| P<sub>228</sub>xxD<sub>331</sub> | W222-D225 | W232-D235 | W222-D225 |
| P676-I685                    | P653-I662 | P667-I676 | P654-I663 |
| I247-F264                    | L241-E258 | I251-V268 | L241-E258 |
| N102-I113                    | C96-S107 | C106-V117 | C96-S107 |
| L611                         | Q595   | L609   | H596   |
| A<sub>665</sub>RR<sub>666</sub> | K643-D645 | G657-V659 | Q644-D646 |
| A240-N243                    | N234-N237 | N244-N247 | N234-N237 |
| P561                         | G546   | G559   | G546   |
| T362                         | C345   | T358   | S645   |
| Q365                         | R348   | R361   | R348   |
| E368                         | D351   | E364   | D351   |
| E369                         | F352   | F365   | F352   |
| E261                         | P225   | D265   | S225   |
| E262                         | G226   | A266   | G226   |
| D271                         | -      | G275   | -      |
| D272                         | -      | T276   | -      |
| Q56-V69                      | E48-F62 | D52-L66 | E48-F62 |
| E261-E276                    | P225-K266 | D265-A280 | S225-K266 |
| D58-G62                      | K50-E55 | Q54-E59 | D50-E55 |
| Q365-E369                    | R348-F252 | R361-F365 | R348-F352 |
| M565-M566                    | W550-Y551 | S563-C564 | W550-Y551 |
| F234-R238                    | F228-F232 | F228-F242 | F228-F232 |
| K587                         | A571   | A585   | V572   |
| I331                         | I316   | I331   | I316   |
| P328                         | P313   | P328   | P313   |
| H372                         | H355   | H368   | H355   |
| I660                         | I637   | I651   | I638   |
| R663                         | R640   | R654   | R641   |
| N664                         | N641   | N655   | N642   |
| D483                         | D463   | D476   | D463   |

<sup>(a)</sup> PlloxA residues highlighted in Figs 2-5 and their counterparts in human LOXs 15LO1, 15LO2 and LOX12. Those fully conserved are highlighted in yellow.
Supplementary References

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