L-amino acid oxidase (LAO) is an enzyme which converts particular L-amino acids into keto acids, ammonia, and H$_2$O$_2$. We found that LAO was presented in acrosomes of sperm, and LAO knockout male, not female, mice revealed a decrease of their litter size, which indicated that LAO plays an important role in displaying sperm function of reproductive male mice. Sperm collected from LAO KO mice showed a high malformation rate and low viability in in vitro culture. In addition, medium cultured with LAO KO sperm contained a low concentration of H$_2$O$_2$ than that of WT sperm. Though there was no difference in the fertilization ratio of oocytes via in vitro fertilization, Western blot and immunofluorescence analysis showed that the level of phosphorylated tyrosine proteins, which is associated with sperm capacitation, was lower in LAO KO sperm than WT sperm. When the LAO KO sperms were cultured with H$_2$O$_2$ supplement, the level of tyrosine protein phosphorylation was increased with a dose dependent manner and the sperm viability was also increased after H$_2$O$_2$ treatment. Those results suggested that sperm LAO provides H$_2$O$_2$ to increase the sperm viability and to induce the capacitation by stimulating the tyrosine phosphorylation levels. Thus, sperm expressing LAO have more chance to reach oocyte at oviduct, which influence the litter size in mice. Moreover, the positive correlation between LAO expression and sperm tyrosine phosphorylation were also found in pig sperm and high expression of LAO were also found in the cow sperm with high fertilization. In summary, LAO may serve as one novel marker for sperm function in different species.