

## Metabolic Engineering of L-Valine Production *Escherichia coli* W3110 and Its Combined Transcriptome and Fluxome Analysis

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### Abstract

We constructed L-valine production strain with *E. coli* W3110 by targeted genetic modification and identified the effect of the biosynthesis of L-valine on cell physiology by combined transcriptome and fluxome analysis. The L-valine-producing strain was constructed by releasing two regulatory mechanisms, feedback inhibition and attenuation. Two amino acids alterations were introduced into *ilvH* which is subject to feedback inhibition by using site-directed mutagenesis. The leader region of *ilvGMEDA* and *ilvBN* operon which is involved in attenuation was changed with the strong *tac* promoter by homologous recombination. *ilvA* was deleted for the prevention of L-isoleucine synthesis resulting in increased pyruvate availability for L-valine biosynthesis. Further improvement of the L-valine-producing strain was achieved by knocking out *leuA* and *panB* thus making more ketoisovalerate available for the L-valine biosynthesis. With this strain 21.5mM L-valine was accumulated within 18 hours of batch fermentation. Combined transcriptome and fluxome analysis during the biosynthesis of L-valine reveals that an increased pyruvate and ketoisovalerate availability is essential to direct the flux into the L-valine biosynthesis. Furthermore, target genes for further metabolic engineering can be selected from the combined analysis data.

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### References

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