Recent studies have demonstrated that xylo-oligosaccharides (XOS), which are classified as emerging prebiotics, selectively enhance the growth of bifidobacteria in general and of Bifidobacterium animalis subsp. lactis strains in particular. To elucidate the metabolism of XOS in the well-documented and widely used probiotic strain B. animalis subsp. lactis BB-12, a combined proteomic and transcriptomic approach was applied, involving DNA microarrays, real-time quantitative PCR (qPCR), and two-dimensional difference gel electrophoresis (2D-DIGE) analyses of samples obtained from cultures grown on either XOS or glucose. The analyses show that 9 of the 10 genes that encode proteins predicted to play a role in XOS catabolism (i.e., XOS-degrading and -metabolizing enzymes, transport proteins, and a regulatory protein) were induced by XOS at the transcriptional level, and the proteins encoded by three of these (beta-D-xylosidase, sugar-binding protein, and xylose isomerase) showed higher abundance on XOS. Based on the obtained results, a model for the catabolism of XOS in BB-12 is suggested, according to which the strain utilizes an ABC (ATP-binding cassette) transport system (probably for oligosaccharides) to bind XOS on the cell surface and transport them into the cell. XOS are then degraded intracellularly through the action of xylanases and xylosidases to D-xylose, which is subsequently metabolized by the D-fructose-6-P shunt. The findings obtained in this study may have implications for the design of a synbiotic application containing BB-12 and the XOS used in the present study.