Pyridoxal 5'-phosphate (PLP) biosynthesis has been studied extensively in *Escherichia coli* K-12 by a combination of biochemical and genetic approaches, and the *E. coli* pathway serves as a benchmark for understanding PLP biosynthesis in other organisms. The goal of this brief review is to highlight some of the genomic and genetic approaches that have contributed to delineating the *de novo* and salvage pathways of PLP biosynthesis in *E. coli*. Classical genetic approaches provided the initial *pdx* mutants, and the combination of genomics and genetics has helped to elucidate the functions and redundancy of enzymes involved in PLP biosynthesis. Genetic approaches have also verified intermediates in the PLP biosynthetic pathway in bacterial cells.

Pyridoxal 5'-phosphate (PLP) is the active form of vitamin B₆ and acts as an essential, ubiquitous coenzyme in many aspects of amino acid and cellular metabolism (see [1,2]). PLP is synthesized *de novo* in *E. coli* by converging branched pathways that lead to 4-phosphohydroxy-L-threonine (4PHT) and D-1-deoxyxylulose phosphate (DXP), which are condensed to form pyridoxine 5'-phosphate (PNP) (Fig. 1) [2-9]. PNP is then oxidized by PdxH oxidase to form PLP, the active coenzyme (Fig. 1) [1,2,10,11]. PLP is converted to pyridoxamine 5'-phosphate (PMP) by the half-reaction of transaminases (Fig. 1) (see [1,2,6]). PMP is recycled back to PLP by the second half-reaction of transaminases and by PdxH PNP/PMP oxidase in the PLP salvage pathway (see below). Important new findings suggest that part or all of the *de novo* pathway for PLP biosynthesis is different in fungi, plants, and some bacteria from that in *E. coli* [12,13].

PLP can also be synthesized in *E. coli* by a salvage pathway that utilizes pyridoxal (PL), pyridoxine (PN), and pyridoxamine (PM) taken up from the growth medium (Fig. 2).
Figure 1. *de novo* PLP biosynthetic pathway in *E. coli* K-12. See text for details.

[10,11,14,15]. In the salvage pathway, PL, PN, and PM are first phosphorylated by the PdxK PN/PL/PM kinase or the PdxY PL kinase to form PLP, PNP, and PMP, respectively (Fig. 2) [14,15]. PNP and PMP are oxidized by the PdxH oxidase, which functions in both the salvage and *de novo* pathways [10,11]. Similar salvage pathways are present in other microbes and in mammalian cells that lack a *de novo* PLP biosynthetic pathway (see [10,11,14,15]). In mammalian cells, PLP homeostasis is further maintained by the offsetting activities of PL kinases and a PLP-specific phosphatase (P'ase) [16]. A cytoplasmic PLP P'ase activity has been detected in *E. coli* K-12, but it has not yet been determined whether this P'ase is specific for PLP [Y. Yang and M. Winkler, unpublished result].

Extensive early hunts for mutants of *E. coli* B or *E. coli* K-12 that require PN or PL for growth were carried out using classical methods, such as antibiotic enrichment, by Walter Dempsey and coworkers (see [1,2]). The *pdx* biosynthetic genes were grouped into five different linkage groups, which later complementation studies showed each consist of a single distinct gene [17]. *pdxB* mutants of *E. coli* K-12 require PN, PL, or glycolaldehyde (CHO-CH₂OH) (GA)