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Amino-sugar Transport Systems of *Escherichia coli* K12

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Glucosamine, mannose and 2-deoxyglucose enter *Escherichia coli* by the phosphotransferase system coded for by the gene *ptsM*. The glucosamine- and mannose-negative, deoxyglucose-resistant phenotype of *ptsM* mutants can be suppressed by a mutation mapping near *ptsG* that allows constitutive expression of the glucose phosphotransferase coded for by the gene *ptsG*.

N-Acetylglucosamine enters *E. coli* by two distinct phosphotransferase systems (White, 1970). One of these is the PtsM system, the other is coded for by a gene which maps near the *nagA,B* genes at about min 15 on the *E. coli* chromosome. We propose that this gene be designated *ptsN*. Strains with either of these components of the phosphotransferase system will utilize *N*-acetylglucosamine as sole carbon source.

INTRODUCTION

Many, though not all, sugars enter *Escherichia coli* by a phosphotransferase mechanism (Kundig *et al.*, 1964). For transport of a sugar to occur in this manner the phosphate group of phospho*enol*pyruvate is transferred, in a reaction catalysed by Enzyme I, to a small histidine-containing protein, HPr. The phosphate group is then transferred from the phosphorylated HPr to the appropriate sugar, which appears inside the cell as the sugar phosphate. The latter, sugar-specific, transfer reaction is catalysed by one of a series of enzymes generically known as Enzymes II. However, the specificity for their sugar substrates exhibited by Enzymes II is not absolute (for review, see Kornberg, 1976).

The gene *ptsM* [originally called *mpt* by Curtis & Epstein (1975) and *ptsX* by Kornberg & Jones-Mortimer (1975)] codes for such an Enzyme II. This Enzyme II (PtsM system) effects the transport of a number of hexoses with the D-*arabino* configuration. Thus, Curtis & Epstein (1975) have shown that glucose, mannose, glucosamine, 2-deoxyglucose and *N*-acetylmannosamine are all substrates for this system and we (Kornberg & Jones-Mortimer, 1975) have shown that fructose is too. In the present paper, we show that one of the two phosphotransferases for *N*-acetylglucosamine described by White (1970) is that specified by *ptsM* and that the other phosphotransferase for *N*-acetylglucosamine maps (as White predicted) near the genes for *N*-acetylglucosamine-6-phosphate deacetylase (*nagA*) and glucosamine-6-phosphate deaminase (*nagB*), at about min 15 on the *E. coli* chromosome. We propose that the gene for this phosphotransferase be described by the symbol *ptsN*.

We suggested (Kornberg & Jones-Mortimer, 1975) that glucosamine might also enter *E. coli* by the PtsG system (formerly described as Umg or Gpt), the major phosphotransferase for glucose, provided that this system was expressed constitutively or had previously been induced. We have confirmed this suggestion by selecting PtsG-constitutive mutants as spontaneous glucosamine-positive derivatives of a *ptsM* strain, and by showing that glucose-negative derivatives of such a strain (*ptsG*) become simultaneously glucosamine-negative (and mannose-negative) and deoxyglucose-resistant.

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Table 1. *Strains of Escherichia coli used*

All strains are Hfr PO2A, except K1.1.4 which is PO1, and 5K-C600 and HK360 which are F⁻.

Strain	Genotype	Reference or source
HK360	*purB ptsG ptsM ptsF umgC*	Laboratory stock
DF1071.2B	*eda*	Faik *et al.* (1971)
K1.1.4	*thyA metB pps gltA*	Laboratory stock
5K-C600	*thr leu lac hsdR supE rps*	S. Glover (Jones-Mortimer, 1973)
JM1077	*thyA hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK ptsF*	Henderson *et al.* (1977)
JM1080	*thyA hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK ptsF galP*	JM1077 (Henderson *et al.*, 1977)
JM1110	*thyA hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK ptsF galP umgC*	JM1080 (Glucosamine^{-})
JM1118	*thyA hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK ptsF galP umgC ptsG*	JM1110 (Glucose^{R} on glycerol)
JM1120	*thyA hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK ptsF ptsN*	JM1077 (*N*-Acetylglucosamine^{R} on glycerol)
JM1121	*thyA hisgnd*$^{\Delta}$ *fda*^{ts} *mglP galK ptsF ptsN eda*	P1.DF1071.2B × JM1120; Glucosamine^{+}
JM1123	*thyA hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP ptsF ptsN gltA*	P1.K1.1.4 × JM1120; Galactose^{-}
JM1138	*thyA hisgnd*$^{\Delta}$ *fda*^{ts} *mglP galK ptsF galP umgC ptsG eda*	P1.DF1071.2B × JM1118; Glucosamine^{+}
JM1141	*thyA hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK ptsF galP umgC ptsG eda*	JM1138 (Glucosamine^{R} on glycerol)
JM1156	*thyA hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK ptsF galP umgC ptsN*	JM1110 (*N*-Acetylglucosamine^{R} on glycerol)
JM1111	*cysI hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK uhpC srlA*	from JM1077 in four stages
JM1158	*cysI hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK uhpC srlA ptsN*	JM1111 (*N*-Acetylglucosamine^{R} on glycerol)
JM1161	*cysI hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK uhpC srlA supE*	P1.5K-C600 × JM1158; *N*-Acetylglucosamine^{-}
JM1165	*cysI hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK uhpC srlA supE ptsN*	JM1161 (*N*-Acetylglucosamine^{R} on glycerol)
JM1166	*cysI hisgnd*$^{\Delta}$ *fda*^{ts} *ptsM mglP galK uhpC srlA supE ptsN*	
JM1179	*thr leu lac hsdR supE ptsN rps*	5K-C600 (2-Deoxy-2-iodoacetamidoglucose^{R} on glucose)

METHODS

Cultures were grown on the minimal medium of Ashworth & Kornberg (1966) or on Oxoid nutrient broth (some batches of which proved to inhibit the strains). Growth rates were determined using an EEL nephelometer. The uptake of ^{14}C-labelled sugars (from a concentration of 50 μM) was measured by a modification of the method of Kornberg (1972) in which the sugar was added to cultures growing exponentially with lactate as the carbon source at 20 °C. P1-mediated transduction was carried out by the method of Lennox (1955). The strains of *E. coli* employed are listed in Table 1. The genetic symbols are those of Bachmann *et al.* (1976), except that the additional symbols *ptsN* and *umgC* are used to indicate genes for the *N*-acetylglucosamine phosphotransferase and for the constitutive expression of the PtsG glucose phosphotransferase, respectively. The bacteriophage T4 mutant N82 was a gift from Dr E. McFall, and 2-deoxy-2-iodoacetamido-D-glucose from Dr P. W. Kent.

RESULTS AND DISCUSSION

Entry of glucosamine into mutants constitutive for the PtsG system

About 10^8 cells of strain JM1080 were plated on solid medium with glucosamine as sole carbon source and incubated at 30 °C for 3 d. Since this strain is *ptsM* it does not grow on glucosamine unless the PtsG system is induced. Glucosamine is, however, not an inducer of that system: glucosamine-positive colonies are therefore likely to form PtsG constitutively. Single-colony isolates were made from colonies that appeared on this medium and tested for their ability to grow on solid medium with glucosamine or mannose as carbon source and to take up methyl α-D-[^{14}C]glucoside from nutrient agar plates also containing that glucose analogue at 10 μM. One strain, JM1110, which had the desired properties was chosen for subsequent experiments. That strain JM1110, which was shown to be *umgC* by direct

Table 2. *Mean generation times of E. coli strains on different carbon sources*

Carbon sources were present at 10 mM, except lactate which was at 20 mM. Mean generation times are expressed in h.

		Carbon source					
Strain	Genotype	Mannitol	Glucose	Glucos-amine	Mannose	Lactate	Lactate + 2-deoxy-glucose
JM1080	*ptsM ptsG*$^+$*umgC*$^+$	2·0	2·0	>48	7	2·3	2·3
JM1110	*ptsM ptsG*$^+$*umgC*	2·2	2·2	2·2	2·2	2·9	8·2
JM1118	*ptsM ptsG umgC*	1·9	>24	>24	>24	3·0	3·0
JM1138	*ptsM*$^+$*ptsG umgC*	2·5	>48	3·5	3·0	2·6	7·0

Table 3. *Uptake of methyl* α-D-[^{14}*C*]*glucoside and 2-deoxy*[^{14}*C*]*glucose by E. coli strains*

			Uptake [nmol (mg dry wt)$^{-1}$ min^{-1}]	
Strain	Genotype	Carbon source	Methyl α-glucoside	2-Deoxy-glucose
JM1080	*ptsG*$^+$*ptsM umgC*$^+$	Lactate	1·8	<0·1
JM1080	*ptsG*$^+$*ptsM umgC*$^+$	Glucose	8·4	ND
JM1110	*ptsG*$^+$*ptsM umgC*	Lactate	9·3	0·4
JM1118	*ptsG ptsM umgC*	Lactate	<0·01	0·3
JM1138	*ptsG ptsM*$^+$*umgC*	Lactate	0·03	31

ND, Not determined.

assay of methyl α-D-glucoside uptake, is still *ptsM* (that is, that it carries a mutation suppressing the glucosamine-negative phenotype of strain JM1080 rather than a reverted *ptsM*$^+$ allele) was further demonstrated as follows. Bacteriophage P1 was propagated on the strain and used to transduce strain JM1121 (*eda ptsM*$^+$*ptsN*) to Eda$^+$, selecting for recombinants able to grow on gluconate as sole carbon source. Of 44 such recombinants, 50% had lost the ability to grow on glucosamine, and must therefore have acquired the *ptsM* allele of the donor strain since *ptsM* and *eda* are cotransducible at about this frequency (Jones-Mortimer & Kornberg, 1974).

Since our previous results (Kornberg & Jones-Mortimer, 1975) had indicated that a constitutive PtsG system might transport glucosamine, two types of experiment were carried out to demonstrate the involvement of the PtsG system. A mutant, strain JM1118, was isolated from strain JM1110 as being resistant to glucose whilst growing on glycerol at 41 °C, by the appropriate modification of the method of Jones-Mortimer & Kornberg (1976). This strain had simultaneously lost the ability to grow on glucosamine and mannose, as is shown in Table 2, which compares the growth of this strain with strains JM1110, JM1080 and JM1138 on glucose, mannose, glucosamine and mannitol, and on lactate in the presence and absence of 2-deoxyglucose. The properties of strain JM1118 show that it is *ptsG* as well as *ptsM* and indicate that its parent, strain JM1110, uses the PtsG system for the transport of glucosamine, mannose and 2-deoxyglucose.

Strains JM1080, JM1110, JM1118 and JM1138 were assayed for the presence of methyl α-D-glucoside and 2-deoxyglucose transport systems during growth on lactate (when the system should not be induced). Strain JM1080 was also assayed for the methyl α-D-glucoside transport system after growth on glucose. The results of these experiments (Table 3) show that strain JM1118 lacks the methyl α-D-glucoside transport system, whereas induced cells of strain JM1080 and uninduced cells of strain JM1110 each have about five times as much transport system activity as uninduced cells of strain JM1080. Of these strains only JM1138

(*ptsM*$^+$) takes up significant quantities of 2-deoxyglucose, even though it fails (being *ptsG*) to take up methyl α-D-glucoside.

The lesion in strain JM1110 was mapped in the following way. Strain HK360 (*ptsG umgC ptsM purB*) was transduced with P1 grown on strain DF1071.2B, which forms the PtsG system inducibly, and glucose-positive recombinants were selected. Of 36 of these examined, nine were constitutive for PtsG as judged by the film technique of Kornberg (1972): these grew with mannose or glucosamine as carbon source. The other 27 recombinants were inducible for PtsG and unable to utilize either mannose or glucosamine. (No PtsM$^+$ recombinants were observed in this sample.) One of these *ptsG*$^+$*umgC*$^+$ recombinants which had retained the *purB* marker was transduced with P1 grown on strain JM1110 and Pur$^+$ recombinants were selected. Of 160 scored, 16% were glucosamine-positive. These results show that the lesion in strain JM1110 is linked to *ptsG* and *purB*, and thus that it maps in about the same position as the PtsG-constitutive allele described by Kornberg & Smith (1972).

These experiments confirm our previous conclusion that glucosamine and mannose can enter *E. coli* by the PtsG system, but are not capable of inducing the system. Though strain JM1110 (*ptsG*$^+$*ptsM umgC*) does not take up significant amounts of 2-deoxyglucose when the extracellular concentration is 50 μM (Table 3), this strain is as effectively inhibited by 10 mM-2-deoxyglucose as is its *ptsG ptsM*$^+$ derivative (Table 2).

Kornberg & Smith (1972) isolated a *ptsG* mutant by selecting for resistance to 2-deoxyglucose with fructose as sole carbon source. On the basis of the specificities of the PtsG and PtsM systems, Postma & Roseman (1976) challenged this procedure and suggested that the selection used should not yield *ptsG* mutants. The strain in which Kornberg & Smith (1972) selected their mutant is known to express the PtsG system constitutively (Kornberg & Reeves, 1972*a*, *b*). Fructose, like 2-deoxyglucose, is a substrate for the PtsM transport system (Jones-Mortimer & Kornberg, 1974; Kornberg & Jones-Mortimer, 1975) and must therefore be a competitive inhibitor of 2-deoxyglucose translocation by this system. Our results (Table 2) confirm that the selection employed by Kornberg & Smith (1972) can yield *ptsG* mutants.

That the inability of strain JM1138 (*ptsG ptsM*$^+$*eda*) to grow on glucose is not the result of glucose being unable to enter the cells is clear from the observation that glucose inhibits the growth of the strain on plates with glycerol as carbon source. A *ptsM* mutant, strain JM1141, derived from strain JM1138 by selection for resistance to glucosamine on medium with glycerol as the carbon source at 41 °C, is also resistant to glucose. We suggest that the reason why strain JM1138 is glucose-negative is that, in strains with a temperature-sensitive fructose-1,6-bisphosphate aldolase, this enzyme limits the rate of growth of the strain (Cooper, 1975) and thus leads to high intracellular concentrations of hexose phosphates (in the case of glucose, particularly of glucose 6-phosphate); this consequently leads to a flux through the hexose monophosphate shunt and the dehydratase of the Entner–Doudoroff pathway. Since the strain carries the *gnd* and *eda* markers, and thus lacks 6-phosphogluconate dehydrogenase and 2-keto-3-deoxy-6-phosphogluconate aldolase activities, 2-keto-3-deoxy-6-phosphogluconate may accumulate. Furthermore, accumulation of 6-phosphogluconate will itself tend to inhibit phosphoglucoisomerase (Muramatsu & Nosoh, 1971) and so exacerbate the situation.

Comparison of the growth rates of strains JM1077 and JM1080 with glucosamine as sole carbon source (Tables 2 and 4) suggests that the gene *galP* also plays a minor role in glucosamine catabolism. Strain JM1077 differs from strain JM1080 only in being *galP*$^+$ (Henderson *et al.*, 1977); it grows on glucosamine poorly but its *galP* derivative does not grow at all. On solid medium the growth of strain JM1077 with glucosamine as sole carbon source is not stimulated by the addition of 1 mM-D-fucose, a compound which is known to induce the GalP transport system to about 10 times the basal level (Henderson *et al.*, 1977). This suggests that although the rate of glucosamine uptake may thus be stimulated, the rate of

Table 4. *Mean generation times of E. coli strains on different carbon sources*

Carbon sources were present at 10 mM. Mean generation times are expressed in h.

		Carbon source		
Strain	Genotype	Mannitol	Glucos-amine	*N*-Acetyl-glucosamine
JM1077	*ptsM ptsN*$^+$	2·3	9	1·9
JM1120	*ptsM ptsN*	2·0	13	>48
JM1121	*ptsM*$^+$*ptsN*	1·8	2·5	2·4

phosphorylation of glucosamine taken up by this route, presumably by a soluble, ATP-dependent kinase as described by White (1968), may be limiting the rate of growth under these conditions. Roehl & Vinopal (1976) have shown that mannose may also enter *E. coli* by the GalP transport system.

Phosphotransferase systems for N-acetylglucosamine

Preliminary experiments indicated that though *N*-acetylglucosamine-negative mutants could readily be isolated, by the appropriate modification of the method of Jones-Mortimer & Kornberg (1976), from *fda*^{ts} strains that were also *ptsM*, they could not be isolated when such strains were *ptsM*$^+$. The system appeared analogous to the one we have previously described (Jones-Mortimer & Kornberg, 1974) where *fpk pfk* double mutants are unable to mutate spontaneously to resistance to 20 mM fructose on glycerol since two independent mutational events would be required. This suggested that the PtsM system might be identical to one of the phosphotransferase activities described by White (1970) as capable of transporting *N*-acetylglucosamine.

In order to test this hypothesis, we isolated from the *fda*^{ts}*ptsM* strain JM1077 a mutant (strain JM1120) which was resistant to *N*-acetylglucosamine at 41 °C and was unable to grow at 30 °C with *N*-acetylglucosamine as sole carbon source. Table 4 compares the growth of this strain with that of its parent. Although both grow well on mannitol and both grow poorly on glucosamine, only strain JM1077 grows readily on *N*-acetylglucosamine.

When bacteriophage P1 grown on strain DF1071.2B (*ptsM*$^+$*eda*) was used to transduce strain JM1120 to *ptsM*$^+$, 30% of the recombinants selected on glucosamine were gluconate-negative and had thus inherited the *eda* allele from the donor strain. All recombinants from this cross were *N*-acetylglucosamine-positive; one which was also gluconate-negative (strain JM1121, Tables 1 and 4) was used in an experiment described earlier in this paper. This experiment, the transduction of *ptsM* from strain JM1110 into strain JM1121 selecting for Eda$^+$ recombinants yielded, as previously stated, glucosamine-negative recombinants. These recombinants proved without exception to be unable to utilize *N*-acetylglucosamine as sole carbon source. These results show that the gene *ptsM* codes for a phosphotransferase capable of transporting *N*-acetylglucosamine and that the gene for the second *N*-acetylglucosamine transport system is not closely linked to *ptsM* and *eda*.

We therefore set out to test White's (1970) suggestion that the gene for the other phosphotransferase (*ptsN*) was linked to the genes *nagA,B* which specify the enzymes for the conversion of *N*-acetylglucosamine 6-phosphate to fructose 6-phosphate. Transduction of strain JM1120 with bacteriophage P1 grown on the *gltA* strain K1.1.4 and selection for galactose-positive recombinants yielded no *N*-acetylglucosamine-positive recombinants among the 385 colonies scored, though 29% of these were *gltA*. Therefore either the *galK* and *ptsN* loci are not cotransducible, or strain K1.1.4 does not carry the *ptsN*$^+$ allele. Preliminary experiments with a recombinant from this cross, JM1123 (*gltA ptsN*), indicated that these genes could be cotransduced, but that neither marker was stable enough

Table 5. *Uptake of N-acetyl[^{14}C]glucosamine by E. coli strains grown with lactate as the carbon source in the presence or absence of N-acetylglucosamine*

Strain	NAG*	Genotype	Uptake of [^{14}C]NAG [nmol (mg dry wt)$^{-1}$ min^{-1}]
JM1077	−	*ptsM ptsN*$^+$	10·2
JM1077	+	*ptsM ptsN*$^+$	12·8
JM1121	−	*ptsM*$^+$*ptsN*	8·8
JM1121	+	*ptsM*$^+$*ptsN*	8·9
JM1120	−	*ptsM ptsN*	1·8
JM1111	−	*ptsM ptsN*$^+$	14·2
JM1161	−	*ptsM ptsN*$^+$	13·2
JM1158	−	*ptsM ptsN*	<0·1
JM1165	−	*ptsM ptsN*	<0·1
JM1166	−	*ptsM ptsN*	<0·1

* *N*-Acetylglucosamine: +, present; −, absent.

to obtain reliable cotransduction frequencies. We therefore attempted to measure the cotransduction between *ptsN* and *supE*. Phage P1 grown on strain 5K-C600 (*supE*) was used to transduce strain JM1158 (*ptsN ptsM cysI*), which is *N*-acetylglucosamine-negative and a cystine auxotroph because of an amber mutation in *cysI*. Of 350 *N*-acetylglucosamine-positive recombinants scored, 102 were glucosamine-positive (i.e. PtsM$^+$) and cystine-negative. Of the remainder, 53 were cystine-negative and 195 grew in the absence of cystine at 30 °C (but not at 40 °C: presumably the suppressed *cysI* gene product is thermolabile). This result indicates either that the cotransduction frequency of *ptsN* with *supE* is 79%, or that the *ptsN* allele in strain JM1158 is suppressible by *supE*. To distinguish between these hypotheses we took a recombinant from this cross, strain JM1161, which was Nag$^+$Cys$^-$ and from it isolated two independent *N*-acetylglucosamine-negative mutants, strains JM1165 and JM1166. When P1 grown on strain JM1077 (*nag*$^+$*supE*$^+$*ptsM*) was used to transduce strains JM1165 and JM1166 to Nag$^+$ the majority of the recombinants tested, 60% and 75% respectively, had lost the ability to grow at 30 °C in the absence of cystine. We therefore conclude that *ptsN* and *supE* are cotransducible. Therefore the gene for the second *N*-acetylglucosamine phosphotransferase maps near *nagA,B*, as predicted by White (1970).

This result was substantiated by the isolation at 40 °C from strain JM1110 (*fda*$^-$*ptsM umgC*) of an *N*-acetylglucosamine-resistant derivative, strain JM1156, which had simultaneously become sensitive to glucosamine at 30 °C. Since this strain, unlike other *N*-acetylglucosamine-negative strains, does not mutate to grow on *N*-acetylglucosamine we consider that it is likely to have a deletion of the genes *ptsN* and *nagB* (the latter specifying glucosamine-6-phosphate deaminase) and possibly also of *nagA* (the gene for *N*-acetylglucosamine-6-phosphate deacetylase). The lesion is cotransducible with *supE*: of 16 *N*-acetylglucosamine-positive transductants obtained with strain 5K-C600 as the donor, 14 were *supE*, as judged by the ability of the T4 amber mutant N82 to form plaques on them.

The results presented in Table 5 confirm that the inability of *ptsM ptsN* double mutants to grow with *N*-acetylglucosamine as sole carbon source (Table 4) is due to a defect in the transport of the sugar, and demonstrate that the amount of sugar transported by either system is sufficient to account for the observed rate of growth. Neither transport system was significantly induced by growth in the presence of *N*-acetylglucosamine.

These results illuminate the finding of White & Kent (1970) and White (1970) that *N*-acetylglucosamine transport-deficient mutants could be isolated by two-stage selection for resistance to *N*-iodoacetylglucosamine, if the first selection was carried out with glucose as the carbon source. Under these conditions the toxic analogue would presumably not be taken up by the PtsM transport system (since glucose also enters by this system) so that

mutants which have lost the *N*-acetylglucosamine transport system should readily be obtained. These mutants, however, will remain sensitive to the toxic analogue during growth on carbon sources which are not substrates for the PtsM system, but resistant mutants selected from such a strain will, by becoming *ptsM*, lose the ability to transport *N*-acetylglucosamine. This hypothesis was tested by isolating from strain 5K-C600 a mutant, strain JM1179, resistant to 2-deoxy-2-iodoacetamidoglucose during growth on glucose, by the method of White & Kent (1970). Bacteriophage P1 grown on this strain was used to transduce strain JM1156, selecting for recombinants on glucosamine as sole carbon source. Of eight transductants analysed all were *N*-acetylglucosamine-negative at 30 °C and resistant to 2-deoxy-2-iodoacetamidoglucose on lactate medium in the presence of glucose. In a control experiment with strain JM1138 as recipient, of 120 Eda^+ recombinants tested, none was resistant to the analogue. Therefore, the conditional analogue-resistant mutants of the type described by White & Kent (1970) have lost the *N*-acetylglucosamine transport system specified by the gene *ptsN*.

Mutants deficient in glucosamine-6-phosphate synthetase (*glmS*) require glucosamine or *N*-acetylglucosamine for growth (Wu & Wu, 1971; Imada *et al.*, 1977). In the presence of glucose only *N*-acetylglucosamine satisfies their growth requirement. Imada *et al.* (1977) suggested that this is because glucose inhibits the phosphotransferase for glucosamine but not that for *N*-acetylglucosamine. Our results indicate that, since glucosamine must enter cells by either the PtsM or PtsG system, glucose should be a competitive inhibitor of both uptake systems for glucosamine, though it need not be an inhibitor of the PtsN system for *N*-acetylglucosamine for which it is not a substrate.

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