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Review

# Gastrointestinal polyamines and regulation of mucosal growth and function

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## Introduction

The polyamines, putrescine, spermidine, and spermine, are a group of ubiquitously distributed organic cations that are intimately involved in proliferation. In eukaryotic cells, the parent polyamine, the diamine putrescine, is synthesized from the amino acid ornithine by the enzyme ornithine decarboxylase (ODC). Sequential addition of two propylamine groups that are supplied by decarboxylated s-adenosylmethionine results in synthesis of the higher polyamines spermidine and spermine (*Fig. 1*). In contrast, prokaryotes express three decarboxylases whose action yields polyamines. In addition to ODC, arginine decarboxylase also yields putrescine and lysine decarboxylase acts in prokaryotes to synthesize cadaverine from lysine. As mammalian cells have retained only one enzyme, ODC, the presence of cadaverine in mammalian tissues serves as a marker for polyamines derived from prokaryotic metabolism. The pKa's of the amino groups of polyamines are in the range of 10 to 11 and at the physiological pH of 7.4, polyamines exist as highly charged polyacations.

All mammalian cells express the enzyme ODC and are able to synthesize polyamines. Cells of the gastrointestinal epithelium, however, are in the unusual position of having both endogenously synthesized polyamines available for metabolism and a large supply of exogenous polyamines available from the gut lumen. The functions of endogenously synthesized polyamines by eukaryotic cells and the relationship of polyamines to cellular proliferation has been the subject of several excellent recent reviews[1,2,3] and will not be discussed further. This article, instead, will concentrate on a review of polyamines in the lumen of the gastrointestinal tract and will be divided into sections describing the putative sources of intraluminal polyamines, their transport across the epithelium, and their potential importance to treatment of tumors both inside and outside the gut. For the purposes of this report, exogenous polyamines will be defined as those available from the lumen of the gastrointestinal tract as opposed to polyamines derived from endogenous, intracellular polyamine biosynthesis by cells of the mucosa.

## Polyamines in the gastrointestinal lumen

The gastrointestinal lumen of the rat contains high concentrations of putrescine.[4,5] In the proximal gut, putrescine concentration is in the millimolar range and falls progressively from duodenum to ileum (*Table 1*). The putrescine is not bound to protein or cell membranes and appears to be free in solution. The lumen contains substantially less spermidine and little to no spermine. In other extracellular compartments such as blood, for example, putrescine concentration is in the submicromolar range.[6] Only semen with a spermine concentration between 5 and 15 mM[7] contains the concentration of polyamines found in the gut lumen. The source of polyamines identified in gut lumen remains in doubt, but may be of prokaryotic origin. The lumen of the distal gastrointestinal tract contains a resident microflora. Additionally, many prokaryotes lack the spermidine and spermine synthase that are required for synthesis of the higher polyamines.[8] The presence of cadaverine in luminal chyme coupled with the low concentrations of spermidine and spermine are suggestive of a prokaryotic source for these amines. Cadaverine was identified by two groups who have measured polyamines in rat intestinal chyme, but the concentrations of cadaverine identified differed by a substantial margin. Polyamines have also been measured in the human digestive lumen using an intestinal perfusion technique. As reported for the rat, high levels of putrescine and cadaverine and low levels of spermidine and spermine were

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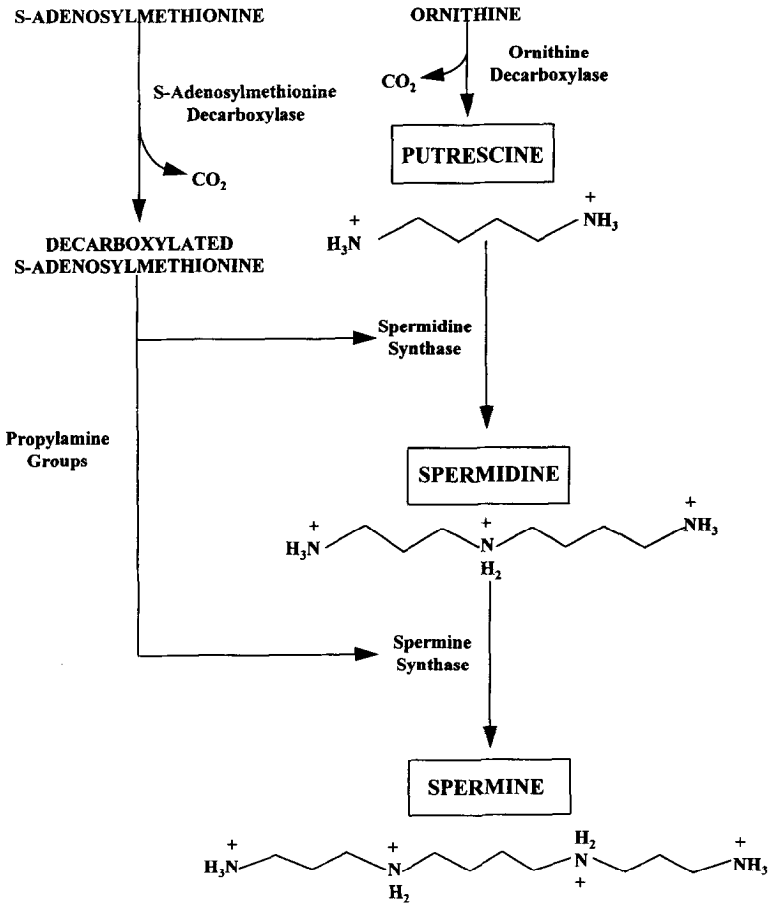


**Figure 1** Biosynthesis of the three primary mammalian polyamines putrescine, spermidine and spermine. Sequential additions of propylamine groups from decarboxylated *S*-adenosylmethionine are used in the synthesis of spermidine and spermine by the enzymes spermidine and spermine synthase.

identified (D. Tome, personal communication, see also Ref. 9). Human bile also contains polyamines.[10,11,12] The polyamine profile of human bile mirrors that of the intestinal lumen with millimolar levels of putrescine and cadaverine and much lower levels of spermidine and spermine.[12] The identification of cadaverine in both bile and chyme suggests these polyamines derive from prokaryotic polyamine biosynthesis and is believed to reflect polyamine metabolism by the intestinal microflora.

Interestingly, chyme collected from duodenal and jejunal lumen, although containing high concentrations of polyamines, contained neither the prokaryotic or eukaryotic enzymes required for polyamine synthesis. In contrast, ileal and colonic chyme displayed both lysine and arginine decarboxylase activity, polyamine biosynthetic enzymes that are exclusive to prokaryotes, suggesting that mixed intestinal contents in the distal gut contain bacteria capable of synthesizing polyamines.[4] The presence of what seem to be polyamines of bacterial origin in bile and small intestinal lumen, which contain few bacteria, presumably reflects enterohepatic recirculation of microfloral synthesized polyamines that have been absorbed into the portal circulation and resecreted into duodenal lumen as a component of bile.[4]

**Table 1** Concentrations of polyamines in the lumen of the gastrointestinal tract of the rat

| | Putrescine | Cadverine | Spermidine | Spermine |
|---|---|---|---|---|
| | | | (μM*) | |
| Stomach | 1048 | 451 | 246 | 28 |
| Duodenum | 3043 | 1152 | 426 | 191 |
| Jejunum | 2216 | 1246 | 198 | 104 |
| Ileum | 314 | 65 | 0 | 0 |
| Colon | 680 | 63 | 88 | 7 |

*Values are taken from Ref. 4, where they were originally reported as nmol/g chyme. They were converted to molar concentrations by assuming 1 g of chyme is equivalent to 1 mL water.

The mammalian diet is primarily cellular in nature and as both plant and animal cells contain millimolar concentrations of polyamines, the diet may also provide polyamines to the lumen of the gut. This potentially important area of nutrition has received little research attention, however. It has been estimated that between 100 μmol[13] and 500 μmol[14,15] of polyamines are ingested daily as a component of the diet. Over 80% of spermidine and spermine seem to be absorbed intact, but only 20% of putrescine absorbed from the lumen escapes metabolism, the remaining 80% being converted to either other polyamines or non-polyamine metabolites.[15] In contrast with the paucity of information on the polyamine content of complete diets, the polyamine concentration of mammary gland milk and of a number of pediatric enteral feeding formulations is available. In general, human milk contains more spermidine and spermine (3 to 10 μM) than putrescine (<1 μM); in rat milk, spermidine levels tend to exceed micromolar concen-

trations, whereas both spermine and putrescine are less than 1 μM.[16,17,18] The polyamine content of most pediatric formulas fell in the low micromolar range, with the exception of semi-elemental diets, which had a polyamine profile similar to that of human milk.[18] The investigators speculated that human milk and semi-elemental diets may contain sufficient levels of spermidine and spermine to potentially modulate intestinal maturation in the neonate. Thus, in contrast with the gut lumen, which contains high putrescine and cadaverine concentrations, mammary gland milk and formulas tend to have high spermidine and spermine levels, but contain no cadaverine and little putrescine.

Regardless of the source, it is apparent that the lumen of the gastrointestinal tract contains polyamines in a concentration that is as much as 10,000 times that of other extracellular compartments such as blood. Whether or not luminal polyamines exert a biological action is a current area of investigation.

### *Exogenous polyamines and mucosal function*

The gastrointestinal epithelium is in a constant state of renewal. A stem cell population in the mucosal crypts provides a set of pluripotential daughter cells that can migrate up the villi or deeper into the crypts. As cells migrate, they undergo terminal differentiation into one of the four main cell types that comprise this epithelium; the absorptive enterocyte with its array of brush border digestive enzymes, the mucous secreting goblet cells, the enteroendocrine cells, and the Paneth cell. As the small intestinal lumen contains free putrescine and cadaverine, these cells are chronically bathed in high concentrations of what are potentially biologically active amines. Thus, polyamines have the potential to affect the mucosal proliferative status, the state of differentiation, and finally the function of these four cell types.

The earlier experiments in this field concentrated on determining the effects of luminal polyamines on mucosal proliferation and architecture. Although the lumen of the duodenum and jejunum contain a high concentration of free putrescine, substantially lower amounts are contained in the ileal lumen (*Table 1*). Infusion of putrescine into the ileal lumen of fasted rats resulted in mucosal growth. A 66-hr infusion of putrescine at the rate of 1 μmol/hr produced a 150% increase in ileal mucosal cellularity.[19] Oral administration of 8 μmol of spermine also modulated intestinal cellularity. Thirty hours after oral spermine feeding, an increase in the amount of DNA and weight of small intestine was observed.[20]

Intraluminal polyamine levels were also modulated in experiments in which an intestinal obstruction was introduced by placing an occluding ligature around the intestine. The gastrointestinal mucosa immediately proximal to an intestinal obstruction becomes hyperplastic. Because mucosa that is distal to the obstruction atrophies, it seems the adaptational response to obstruction is regulated by local factors. This model was used to examine the potential contribution of luminal polyamines to the local growth of mucosa surrounding a colonic obstruction.[21,22] In control rats, tissue proximal to an obstruction became hyperplastic and hypertrophic, whereas that distal to obstruction atrophied. Mucosal growth was accompanied by an increase in mucosal spermidine levels that preceded any increase in mucosal (eukaryotic) polyamine biosynthetic activity. On the other hand, prokaryotic polyamine biosynthesis in the colonic lumen proximal to the obstruction was massively induced suggesting the increase in mucosal spermidine derived from prokaryotic metabolism.[22] In a second group of animals, the gut lumen was decontaminated and intraluminal polyamines were depleted before introduction of obstruction by chronic oral gavage with a mixture of nonabsorbable antibiotics to lyse colonic microflora. In these animals, colonic obstruction did not increase luminal polyamine biosynthesis and did not induce mucosal hyperplasia. These data suggest that local, luminal polyamines modulate mucosal growth.

Whether these changes in mucosal proliferative state were direct effects of polyamines independent of other trophic stimuli or simply a permissive action mediated indirectly by polyamine-induced release of systemic trophic factors was addressed in cells in culture. The rat, duodenal, epithelial cell line IEC-6 was cultured in the presence or absence of putrescine.[23] Putrescine treatment of serum deprived cells resulted in a dose-dependent increase in DNA synthesis indicating the polyamine-stimulated cell cycle transition from the $G_0/G_1$ interface into S phase. The magnitude of the response was greater than 75% of that produced when 5% fetal bovine serum (FBS) was used as the mitogen. In addition to increased DNA synthesis, the rates of both RNA and protein synthesis were increased. High performance liquid chromatography analysis of intracellular polyamine levels indicated a selective increase in intracellular putrescine with no change in the concentration of either spermidine or spermine. Cells cultured from tissues other than the gut are also sensitive to treatment with exogenously supplied polyamines,[24,25] suggesting that polyamines may have direct effects on cellular proliferation.

Polyamines may also modulate ontological differentiation of gut epithelium in neonates. Enterocytes in the neonatal rat primarily express lactase activity for digestion of mammary gland lactose during the first 2 weeks of postnatal life, but begin to express an adult pattern of digestive enzyme activity during the third week of postnatal life just before weaning.[26] Intraluminal administration of spermine in suckling rats caused the precocious appearance of sucrase and maltase-isomaltase activity and the disappearance of lactase activity.[20,27,28] The effect was not reproduced when spermine was administered intraperitoneally or when intestinal explants were incubated in spermine-containing solutions, suggesting the polyamine-induced release of systemic factors from the mucosa that mediated the effect. Serum adrenocorticotrophic hormone (ACTH) levels were increased 6 to 7 fold after oral administration of 8 μmol of spermine. Spermine-induced precocious maturation was not observed in adrenalectomized animals. Again, the effect on glucocorticoid release was not reproduced when spermine was administered intraperitoneally.[28] As effects were seen only when spermine was administered by the enteral route, it was concluded that the effects of the polyamine on disaccharidase ontology were mediated by polyamine-induced release of an unidentified gut hormone that increased adrenal ACTH release. Administration of

gastrin, cholecystokinin, glucagon 1-37, or secretin failed to affect dissacharidase activities. Thus, unlike the effects of polyamines on mucosal growth, which may be direct, the effects of polyamines on brush border enzyme maturation seem to be indirect and dependent on polyamine-induced release of as yet unidentified factors from the gut.

Ornithine α-ketoglutarate (OKG) is composed of two molecules of ornithine and one molecule of α-ketoglutarate and improves the nutritional status in protein-depleted patients. Oral administration of OKG in adult rats produced a small, but significant increase in small intestinal villus height and crypt depth and a larger increase in the activities of the brush border digestive enzymes sucrase, lactase, and aminopeptidase.[29] Oral OKG increased mucosal ornithine by 44% and putrescine by nearly 300%. Much of the putrescine was converted to γ aminobutyric acid by intestinal diamine oxidase. Whether the effects of OKG on intestinal morphology and digestive enzyme activities was mediated by putrescine or gamma-aminobutyric acid was not determined, but oral administration of the polyamine precursor ornithine was accompanied by mucosal growth and differentiation.

Three groups have reported that polyamines modulate activity of the brush border glucose transporter in epithelial of mature animals. In the mid 1980s, Meezan and coworkers reported that 50 μM concentrations of either putrescine, spermidine, or spermine-stimulated glucose uptake into rabbit renal brush border membrane vesicles (BBMV).[30] Diffusional glucose uptake was unaffected, indicating that polyamines stimulated the active transport of glucose rather than inducing nonspecific changes in membrane lipid properties. They went on to demonstrate that spermine was incorporated into several brush border membrane proteins ranging in weight from less than 10 kDa to over 200 kDa, suggesting that polyamine effects on glucose transport were caused by covalent modifications of membrane proteins, modifications that may have been catalyzed by transglutaminase. On the other hand, addition of polyamines or their analogs to LLC-PK1 kidney cells in culture down-regulated glucose transport, whereas depletion of polyamines resulted in a 4- to 5-fold increase in glucose transport.[31] Polyamines also affect glucose transport in BBMV prepared from rabbit small intestine.[32] Glucose transport was decreased in BBMV prepared from rabbits treated with difluoromethylornithine (DFMO), a suicide substrate inhibitor of ODC that depletes cells of polyamines. The effect of DFMO was prevented by pretreatment of rabbits with orogastric putrescine, spermidine, or spermine, 10 mL of a 2 μM solution. In rabbits receiving only orogastric spermine, glucose transport was significantly increased. Thus, both in vitro and in vivo treatment with polyamines modulates activity of the brush border membrane glucose transporter.

In summary, exogenously supplied polyamines in concentrations substantially lower than have been reported to exist in situ in gut lumen can modulate mucosal growth, epithelial cell proliferation, ontological development, and activity of brush border proteins. For these highly charged cations to reach the intracellular space would require a membrane transport system to move polyamines from the intestinal lumen across the plasma membrane.

## Polyamine transport systems

The intracellular polyamine pool is regulated not only by the rate of polyamine synthesis and degradation, but also by rate of uptake and release. Uptake systems for the different polyamines have been characterized by kinetic studies in prokaryotes as well as in eukaryotes, but the carriers have been cloned only in prokaryotic systems. Early studies demonstrated that *Neurospora crassa* possesses a polyamine transport system(s) able to transport all three natural polyamines.[33] In *Escherichia coli*, polyamine uptake was energy dependent, and the putrescine transport system was different from the spermidine and spermine transport system[34,35] suggesting a multiplicity of polyamine transport sites.

The existence of multiple transport sites was confirmed when Igarashi and coworkers successfully cloned and characterized three different polyamine transport systems from *E. coli*. One clone (pPT104) codes for a transport system capable of transporting both putrescine and spermidine. This transport system consists of four proteins A, B, C, and D of respective molecular weights 43, 31, 29, and 39 kDa. The A protein is a membrane-associated protein that included a consensus amino acid sequence for a nucleotide binding site. B and C proteins consisted of six putative transmembrane-spanning segments linked by hydrophilic segments of variable length. D protein was suspected to be a polyamine binding protein existing in the periplasmic space.[36] Two other clones (pPT79 and pPT71) code for transport systems catalyzing only the transport of putrescine. The transport system encoded by pPT79 was a periplasmic system consisting of four proteins similar to those encoded by pPT104. In contrast, pPT71 (potE) had a 1,317-nucleotide open reading frame encoding a 439-amino acid protein with 12 putative transmembrane segments.[37] Studies using inside-out membrane vesicles showed that potE protein was able to exchange putrescine and ornithine by an antiport mechanism.[38]

Kinetic analysis of polyamine transport systems in mammalian cells also suggest the presence of multiple transport systems as is the case in bacteria. In human lymphocytes, spermidine, and spermine were taken up by a common transport system that had a much lower apparent affinity for putrescine.[39] Bovine adrenocortical cells and porcine kidney cells (LLC-$PK_1$) possess at least two different transport systems for polyamines; putrescine uptake seemed to be the sum of a sodium-dependent, saturable process and a nonsaturable, sodium-independent component, whereas spermine uptake was mostly sodium independent.[40,41] Similar results demonstrating the presence of multiple transport sites have been obtained with pulmonary alveolar macrophages,[42] human erythrocytes,[43] and type II pulmonary epithelial cells.[44] Using an inhibitor of polyamine synthesis, methylglyoxal bis (guanylhydrazone) (MGBG), and the herbicide paraquat, Byers demonstrated the presence of multiple transport systems in the Chinese hamster ovary cells (CHO).[45] The same group showed that polyamine transport activity was regulated by the intracellular polyamine content in CHO cells. In polyamine-deprived CHO cells, the provision of exogenous polyamines resulted in rapid and large increases in intracellular polyamine content

followed by decreased polyamine transport activity. Protein synthesis was necessary for the increase in transport in response to polyamine depletion.[46] Additionally, expression of a human gene coding for polyamine transport proteins was demonstrated in a polyamine-transport-deficient CHO cell line. Transfection of these cells with human DNA resulted in polyamine transport in the previously transport-deficient cells. Again, kinetic data were consistent with a multiplicity of polyamine-transport systems.[47]

### *Polyamine transport in the gut*

Polyamine transport in tissues of the gut have been studied in whole animals, in acutely isolated epithelial cells, in BBMV prepared from enterocytes, in Ussing chamber preparations, and in cells of gastrointestinal origin in culture. As in other tissues, the data suggest the presence of multiple transport systems.

As mentioned, polyamines are absorbed from duodenum, jejunum, and the colon in the anesthetized rat and seem to recirculate enterohepatically.[4] In addition to absorption from the lumen, it has been suggested that polyamines may be absorbed into small intestinal mucosa from blood.[48,49] In agreement with this suggestion, after intraperitoneal injection of [$^{14}$C]putrescine, significantly more radioactivity was detected in duodenum than in pancreas, liver, kidney, or spleen.[50]

One of the earliest in vitro studies of polyamine transport in the gut was performed in acutely isolated villus enterocytes.[51] Putrescine uptake was saturable, sodium-independent, and inhibited by KCN. Whether polyamines are also transported by cells in the proliferative zone of the crypts was not determined. Examination of polyamine transport in cells of the gastrointestinal tract in culture have yielded qualitatively similar results. In IEC-6, CaCo2, and AR2-4J cells, the $K_m$ for polyamine transport is in the micromolar range.[52,53,54,55] Kinetic analysis of spermidine transport in IEC-6 cells was consistent with the presence of at least two transporters; one site with high affinity ($K_m$ 0.26 μM), but low capacity and a second site with higher capacity but lower affinity ($K_m$ 2.1 μM).[56] Conversely, in colon cancer cells, the three polyamines seem to share a common carrier.[57]

The polyamine transport systems were localized to the apical membrane domain of the enterocyte by employing a model in which the rat duodenal cell line IEC-6 was cultured on raised inserts to promote partial cellular differentiation. Under these culture conditions, the IEC-6 cell expresses tight junctions and begins to segregate apical and basolateral membrane domains.[58] Two classes of apical membrane transporters were identified. One seemed to be nonselective for the three polyamines, whereas the second was specific for those containing aminopropyl groups such as spermidine and spermine. Absorption across the basolateral membrane domain was limited and did not seem to be carrier mediated. Additionally, if cells were preloaded with putrescine, efflux of the polyamines was exclusively into the basolateral compartment.[56] Studies using BBMV prepared from apical membranes of rabbit small intestine confirm the apical membrane localization of the transport sites.[59,60,61] Additionally, polyamine efflux has been measured in inside-out vesicles prepared from basolateral membranes of rabbit enterocytes. As is the case in the apical membrane, efflux across the basolateral membrane domain seemed to be carrier mediated. Using segments of rabbit duodenum, jejunum and ileum mounted in Ussing chambers, Dumontier[62] demonstrated that transepithelial flux of polyamines may differ in different segments of the small intestine. Net putrescine flux was negative in the ileum, but positive in the duodenum suggesting that the latter tissue may be the more important site for putrescine absorption. The absence of net absorption in ileal preparations confirms results in anesthetized rats[4] in which putrescine was not absorbed from the ileum.

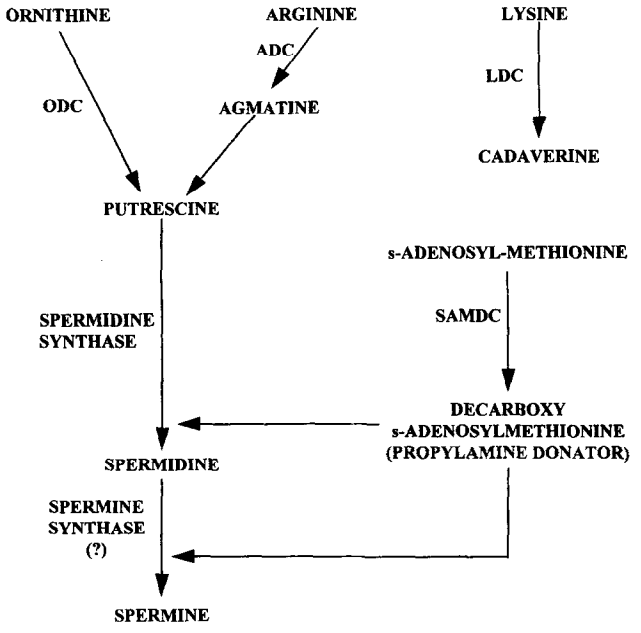


**Figure 2** Biosynthesis of polyamines in prokaryotic cells. The diamines putrescine and cadaverine are synthesized from corresponding amino acids by the enzymes ODC, ADC, and LDC. Only the ODC pathway has been retained in mammalian cells. Some prokaryotic cells lack spermine synthase accounting for the low levels of spermine in some bacteria.

The affinities of the transporter sites for polyamines are in the micromolar range yet are bathed in a polyamine pool that in total is greater than 1 mm. Assuming there is no hindrance to polyamine access to the transport sites, it would appear that under normal physiological conditions the transporters are fully saturated and in a concentration gradient that greatly favors movement of polyamines from the gut lumen into the circulation.

### *Regulation of polyamine transport in the gut*

The primary endogenous regulator of polyamine transport is the protein ODC antizyme.[63,64,65] Antizyme was originally described as an inhibitor of ODC that stimulates the ATP-dependent degradation of ODC by the 26S proteosome. More recently, it has been demonstrated that antizyme is also an endogenous inhibitor of polyamine transport. It is induced during excess polyamine accumulation and thereby plays a unique role to limit polyamine availability by enhancing the rate of ODC degradation as well as

blocking uptake of extracellular polyamines. Antizyme mRNA has been identified, albeit in very low levels in comparison with other tissues, in rat small intestine.[66] We are unaware of other reports of antizyme identification or action in the gut.

In spite of the potential importance of the luminal polyamine pool, regulation of polyamine transport in tissues of the gastrointestinal tract is an area that has received scant attention. Activity of the transporter in cells of the gastrointestinal tract is sensitive to experimental conditions, however, demonstrating that it is under physiological regulation. Polyamine transport rate in the IEC-6 cell is inversely related to rate of cellular proliferation; confluency was accompanied by a 5 fold reduction in $V_{max}$ for putrescine accumulation, 199.5 versus 43.1 pmol/$10^5$ cells-hr, with no change in $K_m$.[67] Additionally, tumor-bearing rats seem to evolve an adaptive response in the small intestinal mucosa involving an increased capacity of the brush border membrane to transport polyamines.[68]

The signals mediating these effects remain to be identified, but both the extracellular matrix and circulating, systemic factors may be involved. The effect at the level of the extracellular matrix may be related to the change in cell shape that accompanies growth density arrest as it can be duplicated by artificially modulating cell shape by plating cells at subconfluent densities on nonadherent matrices.[69] Perhaps surprisingly, however, a number of gut hormones, which are trophic to tissues of the gut, are also negative affectors of polyamine transport.[67,70,71] Gastrin, secretin, and cholecystokinin all reduce the uptake of polyamines into gastrointestinal cells by between 20% and 50%. Conversely, EGF, insulin, and IGF-1-enhanced putrescine uptake into pancreatic acini by 30 to 40%.[71,72] The stimulatory effect of EGF was not sensitive to cycloheximide, suggesting EGF might affect translocation of pre-existing transporters into the membrane.[73] As all of these hormones and growth factors stimulate mucosal and/or pancreatic proliferation, why some enhance and others inhibit polyamine uptake is unknown. Additionally, what intracellular second messenger systems these peptides use to transduce their signals remains a largely unexplored area. An unidentified intracellular signaling pathway involving $Ca^{2+}$/calmodulin may be involved, however. Treatment of the IEC-6 cell with 10 μM putrescine produced an increase in free ionized intracellular calcium raising $[Ca^{2+}]_i$ from a basal level of 112 nM to 313 nM.[67] Measurement of putrescine transport in 0 $Ca^{2+}$/0.5 mM EDTA suggested that an intracellular source of calcium was used during putrescine transport. Buffering the rise in intracellular calcium with BAPTA or pretreatment with the calmodulin antagonist W-7 inhibited polyamine transport. Calcium is also involved in polyamine transport in murine leulemia cells.[74]

## Luminal polyamines and growth of tissues outside the gut

Despite promising results in cell culture models, clinical trials with DFMO as a single agent have not shown lasting antitumor effects in humans. It appears that DFMO inhibition of de novo polyamine synthesis by tumor cells is not sufficient to inhibit proliferation because of the availability of exogenous polyamines to support tumor cell proliferation. The source of exogenous polyamines seems to be the gut. Seiler and coworkers have demonstrated that cancer cells implanted in rats escape polyamine antimetabolite therapy because they are able to recruit polyamines from the large supply of bacterial or dietary derived polyamines in the gut lumen.[75,76] Limiting polyamine availability by placing rats on a polyamine-free diet, coupled with partial decontamination of the gut by treating with nonabsorbable antibiotics significantly decreased the growth rate of a number of implanted solid tumors including Lewis lung carcinomas, fibrosarcomas, prostatic adenocarcinomas, and intracranial glioblastomas.[77,78,79,80,81] In the case of Lewis lung carcinomas, a relatively DFMO-sensitive tumor, tumor cell proliferation was inhibited by 80 to 100% and survival time significantly increased. In the DFMO-insensitive U-251 human glioblastoma xenografted into nude mice, DFMO produced an almost complete inhibition of tumor growth.[82] A second model was tested in which transformed leukemic cells that did not express polyamine transporters was implanted into mice. Survival of wild-type L1210 leukemic mice xenographs was only slightly prolonged by DFMO treatment. Mice xenographed with the L1210-$MGBG^r$ polyamine transport-deficient mutant and treated with DFMO had a 30 to 75% cure rate, again demonstrating that transport of exogenous polyamines can support growth of tumors outside the gastrointestinal tract.[83]

Although a number of models have demonstrated that exogenous polyamines recruited from the gastrointestinal lumen can supplant endogenous polyamine biosynthesis during growth of tumors, no data are available regarding the potential use of exogenous polyamines during growth of normal tissues outside the gut. Although it has not been examined, it is tempting to speculate that normal tissues may also use transport of exogenous polyamines for maintenance of the intracellular polyamine pool. Whatever the case, it is clear that mammalian cells express two separate systems, synthesis and transport, both of which are capable of filling the demand for polyamines. The transport system seems to be at least partially and perhaps totally dependent on polyamines available from the lumen of the gastrointestinal tract.

In summary, the lumen of the gastrointestinal tract contains millimolar concentrations of two polyamines, putrescine and cadaverine. The presence of cadaverine suggests luminal polyamines may be derived from prokaryotic polyamine metabolism although significant amounts may also be supplied by the diet. Exogenous polyamines modulate mucosal proliferation, differentiation, and absorption of diet. They bathe a set of polyamine transport sites with affinities for polyamines that are in the micromolar range. Transport of polyamines across the enterocyte appears to be vectoral as transporters favoring absorption are localized to the apical membrane domain of the enterocyte. Study of the physiological regulation of these transport sites is in its infancy, but deserves further attention as they can without question contribute polyamines to support neoplastic growth and might be involved in maintenance of the intracellular polyamine pool of untransformed tissues outside the gastrointestinal tract.

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