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Uptake, inter-organ distribution and metabolism of dietary putrescine in the rat

Susan Bardócz, Elizabeth L. Hughes, George Grant, David S. Brown, Tracey J. Duguid, and Arpad Pusztai

The Rowett Research Institute, Greenburn Road, Bucksburn, Aberdeen, UK

The movement of a single dose of ^{14}C-putrescine in the lumen of the rat gastrointestinal tract was followed for 3 hours after intragastric intubation. Putrescine progressed in the gut lumen in a wave-like fashion and was absorbed in the small bowel. Maximal uptake was observed at 2 hours; therefore, this time-point was selected to measure the concentration dependency of putrescine uptake by the small intestine and distribution between the vital organs in a wide concentration range (1/10 to 100 times the dietary input). Putrescine uptake by the small bowel was likely to be by passive diffusion, because the absorption was in proportion to input. The fate of putrescine was determined in the plasma, small bowel, liver, and skeletal muscle by measuring the radioactivity of the polyamines, their acetyl derivatives, and amino acids at physiologic concentrations. It appears that approximately 10% of the dietary input reaches the putrescine body pool. (J. Nutr. Biochem. 9:332–338, 1998)

Keywords: putrescine; luminal uptake; small bowel; skeletal muscle; liver; plasma; internal organs

Introduction

The natural polyamines (putrescine, spermidine, and spermine) are flexible polycations, fully charged under physiologic pH. Because they fulfill a number of roles in cellular metabolism, polyamines are usually considered essential for cell growth and proliferation.[1–5] Polyamines are involved in a number of steps of DNA, RNA, and protein synthesis[1–4] and their cellular concentrations vary according to the stage of the cell cycle.[1–5] They are probably involved in mediating the actions of hormones and growth factors. They may also be required in hitherto undetermined cellular processes.

In the past it was believed that polyamines were synthesized in situ in cells as required, because one of the first steps in cell proliferation is the induction of ornithine decarboxylase (ODC; EC 4.1.1.17, the first enzyme of polyamine synthesis), an event that precedes both nucleic acid and protein synthesis.[4,5] More recently, the importance of polyamines from extracellular sources has been recognized,[6–9] with evidence that, as with indispensable amino acids, sufficient polyamines to support cell renewal and growth can be supplied by the diet.[10]

The small intestine has one of the highest proliferative activities and metabolic rates of all tissues in mammals. Because the surface epithelium is renewed every 2 to 3 days, polyamines might be important for the constant cell division in the crypts of the Liebekuhn of the normally functioning gut. Putrescine, spermidine, and spermine exert specific functions in intestinal mucosal physiology, influencing the integrity and growth of epithelial cells.[11,12] They are required for repair of the damaged mucosa[13] and play a regulatory role in epithelial cell function.[11–13] Therefore, the intracellular profile of polyamines must be highly regulated by simultaneous control of their biosynthesis, catabolism, uptake/transport from endogenous sources, and excretion. The gut lumen is the most important extracellular polyamine source in the gastrointestinal tract, providing polyamines[14] from the diet,[15] from bacteria resident in the gastrointestinal tract,[6–9] or from the enterohepatic circulation.[16]

Putrescine plays a central role in epithelial cell metabolism and its levels are tightly regulated. It is formed from ornithine by ODC in mammalian cells,[1–5] but is also produced from arginine in microorganisms and plants.[17] It

Address correspondence and reprint requests to Dr. Susan Bardócz, The Rowett Research Institute, Bucksburn, Aberdeen AB21 9SB, Scotland, UK.
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is the precursor of both spermidine and spermine, although spermidine can be reconverted to putrescine by acetylation/oxidation via the *polyamine retroconversion pathway*.[18,19] Putrescine concentration is also regulated by uptake/transport and excretion.[18] Enterocytes take up putrescine in vitro,[20,21] using an energy-dependent, quabain-insensitive uptake system that is sensitive to changes in intracellular calcium/calmodulin levels,[22,23] and excrete it in free or acetylated form.[18] Putrescine can be oxidized by diamine oxidase (DAO; EC 1.4.3.6; a copper-dependent enzyme[24]) and, via the action of aldehyde dehydrogenase can be metabolized to γ-aminobutyric acid (GABA, a neurotransmitter[19,24,25]) or succinate.[26]

Although luminal putrescine is a growth factor for the gut[27,28] and, in combination with spermidine and spermine, is necessary for its adaptive growth,[29,30] little information is available on the absorption of dietary putrescine by the gastrointestinal tract, its subsequent utilization by the body, and its bioavailability. Therefore, the aim of the present article was to follow the movement of intragastric ^{14}C-putrescine over a 3 hour period in the gut lumen of rats and to quantify its uptake and distribution between the vital internal organs over a wide concentration range. The fate of putrescine was followed in the plasma, small bowel, liver, and the gastrocnemius muscle at physiologic concentrations by tracing the ^{14}C-label in polyamines, their acetyl derivatives, and amino acids.

Materials and methods

All chemicals used for tissue preparation, polyamine standards, and o-phthalaldehyde were obtained from Sigma (Dorset, UK); HPLC reagents were obtained from BDH (Merck House, Leics, UK); 1,4-^{14}C-putrescine.2HCl, uniformly labelled, and NCS tissue solubilizer fluid were from Amersham International PLC (Amersham, UK); and NE 265 scintillation fluid was from NE Technology Limited (Edinburgh, UK).

Animal care

All management and experimental procedures in this study were carried out in strict accordance with the requirements of UK Animals (Scientific Procedures) Act 1986 by staff licensed under this Act to carry out such procedures.

Animals

Thirty-day-old male Hooded-Lister specific pathogen-free rats of the Rowett colony (approximately 80 g) were kept in individual cages at 20° to 23°C, with a 12 hour light/dark period. After weaning, the rats were housed in groups of five and fed stock diet ad libitum for 1 week. Thereafter they were separated into individual cages and given a highly nutritious, semisynthetic diet with lactalbumin (LA; 100 g/kg diet) as an easily digestible protein source, supplemented with all recommended vitamins and minerals.[31] Based on our measurements this diet contained 11 ± 1 nmoles putrescine and 15 ± 1 nmoles spermidine/g. The rats were offered 6 to 8 g diet/day and distilled water ad libitum.

Experiment 1: Determining small intestinal transit time of putrescine

Groups of five rats (85.2 ± 2.1 g) were kept on LA diet for 3 days prior to the experiment. After an overnight fast, each rat was given 1.5 g of the LA diet. This protocol was necessary because the time elapsed between the last meal and sacrifice affects the hormonal balance of the body (insulin, blood glucose, etc.) and also might affect the levels and distribution of putrescine. The diet also was needed as a bolus of food to accompany the ^{14}C-labeled putrescine (8.5 nmol, 2.4×10^6 dpm/rat), which was given by intragastric intubation in phosphate-buffered saline immediately after the meal was eaten (approximately 2 minutes later). For the luminal uptake studies, five animals per time point were sacrificed at 30, 60, 90, 120, 150, or 180 minutes after intubation with halothane overdose and by cervical dislocation of the neck.

Blood samples were collected in heparinized tubes. The small intestines were removed, weighed, clipped, and cut into 5-cm long segments (i_1 to i_{17}). Each segment was washed with 2 mL of ice-cold saline. The stomach, cecum, and colon also were removed and their contents washed out with 5 mL of ice-cold saline. Aliquots of the washings (0.5 mL) were used for counting radioactivity and 1 mL was used for protein measurement by a modified Lowry procedure.[32] The segments of the small intestinal, cecal, and colonic tissues or 100 μL of blood were placed in NE 265 scintillation fluid for 48 hours and then counted. Stomach tissue was digested in NCS tissue solubilizer for 24 hours before counting. Radioactivity accumulated in the body was calculated as the difference between the dose of ^{14}C-putrescine intubated and the radioactivity recovered in the gut lumen.

Experiment 2: Dose dependency of putrescine absorption

As in Experiment 1, rats were kept on LA diet for 3 days to minimize the polyamine content in their gut lumen and then fasted overnight. The next morning they were fed 1.5 g of LA diet and, immediately after this was eaten, groups of five rats were intubated intragastrically with a mixture of cold and labeled putrescine (keeping the ^{14}C/^{12}C ratio equal, 1:250). Groups 1 through 4 were given 26.7, 267, 2670, or 26,700 nmoles of putrescine, respectively. (Normal intake is approximately 267 nmoles/day.) The rats were sacrificed exactly 2 hours later.

The stomach and small and large intestines were removed and washed with 10 mL of ice-cold saline, and the luminal contents (0.5 mL portions) were counted for radioactivity. Label accumulated in the small and large bowels and the gut contents was determined as previously described.[6,15] Blood was collected, and all major internal organs (pancreas, liver, kidney, lung, spleen, and gastrocnemius muscle) were removed. The small bowel, cecal and colonic tissue, pancreas, spleen, and lungs were placed in NE 265 scintillation fluid for 48 hours and then counted. Stomach, liver (approximately 150–200 mg), one of the kidneys, and hind leg gastrocnemius muscles were digested in 1 mL of NCS tissue solubilizer for 24 hours before the radioactivity was measured.

The amount of putrescine incorporated in the rat body was calculated as the difference between the amount intubated and the luminal contents (indirect method). Radioactivity was also measured directly in the internal organs (as described above and in Bardocz et al.[6,15]) and in the carcass (direct method). Label accumulated into the carcass was measured as follows: the bodies were freeze-dried, weighed, and ground. Proportions (200 mg) were digested with tissue solubilizer and counted. To check the recoveries when counting, another 200 mg portion of the same carcass was spiked with known amounts of ^{14}C-putrescine (2000 cpm/sample) and its radioactivity measured.

Tissue preparation for measurement of radioactivity, polyamine analysis, and recovery of the label

The amount of radiolabel originating from putrescine was measured in samples of the small intestine (18 cm section of jejunum,

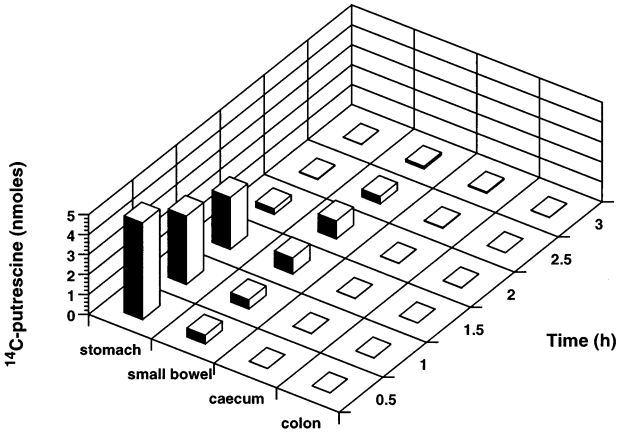

Figure 1 Movements and distribution of ^{14}C-putrescine (nmoles) in the lumen of the gastrointestinal tract (stomach, small bowel, cecum, and colon) at different times (hours) after intragastric intubation of 8.5 nmoles of ^{14}C-putrescine. Data represent means of five rats. Experimental error was approximately ± 5%.

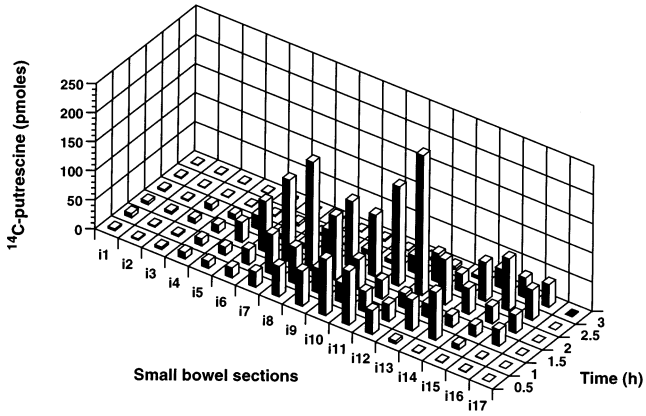

Figure 2 Distribution of ^{14}C-putrescine (pmoles) in the washing of different segments of the small bowel at different times (hours) after intragastric intubation of 8.5 nmoles of ^{14}C-putrescine. Data represent means of five rats. Experimental error was approximately ± 5%.

7–25 cm from the pylorus end), plasma, liver (200–300 mg), and left hind leg gastrocnemius muscle. Tissues were homogenized in 10% ice-cold PCA containing diamino-heptane as internal standard and then diluted to 2% PCA. Plasma (3 mL) was also homogenized in 2% PCA and centrifuged. All tissue and plasma samples were analyzed for polyamines and metabolites by high performance liquid chromatography (HPLC), as described by Seiler and Knödgen.[33] A 450 μL sample (containing 180–3000 dpm) was analyzed by HPLC with the fluorescent detector disconnected. Fractions (45 per sample) were collected and counted for radioactivity. The distribution of radioactivity among the polyamines, their acetyl derivatives, and amino acids was calculated as previously described.[10]

Statistical analysis

The results were subjected to one-way analysis of variance (ANOVA) using the Minitab computer program (Penn State University, State College, PA, USA). When the P-value was less than 0.05, the significance between groups was estimated by Student's *t*-test.

Results

Movement of putrescine in the lumen of the rat gut

The movement of labeled putrescine in the lumen of the rat gut was followed for 3 hours after intragastric intubation (*Figure 1*). By 2.5 hours less than 5% of the label remained in the stomach content. As the putrescine progressed toward the ileum, by 30 minutes it had reached segment 14 of the small bowel, 70 cm from the pylorus, reaching its maximal concentration in the gut lumen by 2 hours (*Figure 2*). Although only tiny amounts of putrescine were detected in the lumen of the terminal ileum (section i_{17}) in the first hour (*Figure 2*), counts originating from putrescine were already present in the luminal contents of the cecum and colon 30 minutes after intragastric intubation (*Figure 1*).

Effect of time on the uptake of putrescine by the small bowel

Thirty minutes after intragastric intubation, radioactivity could be detected in all segments of the small intestinal tissue (*Figure 3*), blood, and internal organs (*Figure 4*). At the times examined, the amounts of label found in each segment of the small bowel tissue correlated with the radioactivity measured in the corresponding luminal washing of the same segment (*Figure 2* and *Figure 3*), giving correlation coefficients of 0.93 to 0.99. The radioactivity measured in the different organs of the body continued to increase for up to 3 hours, but was highest in the blood between 2 and 2.5 hours after intragastric intubation (*Figure 4*). (Because the level of radioactivity in blood was very low compared with other tissues, the values for blood/ml in *Figure 4* are multiplied by a factor of 1000.) The radioactivity absorbed by the gut and accumulated in the body was calculated (indirect method) as the difference between putrescine input and amount recovered in the luminal contents of the gastrointestinal tract at each time point (*Figure 5*). Gut content was minimal 2 hours after intragas-

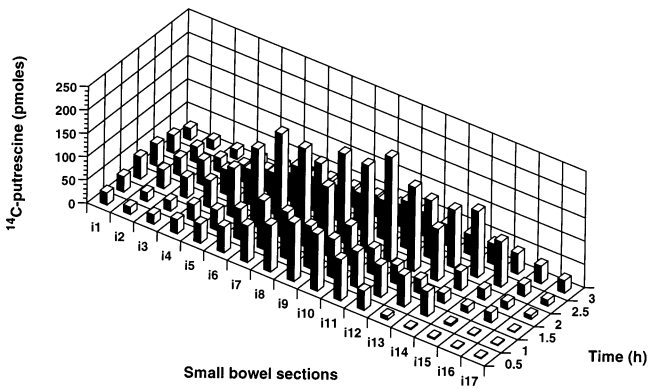

Figure 3 Accumulation of ^{14}C-label (pmoles) in the different tissue segments of the small bowel tissue at different times (hours) after intragastric intubation of 8.5 nmoles of ^{14}C-putrescine. Data represent means of five rats. Experimental error was approximately ± 5%.

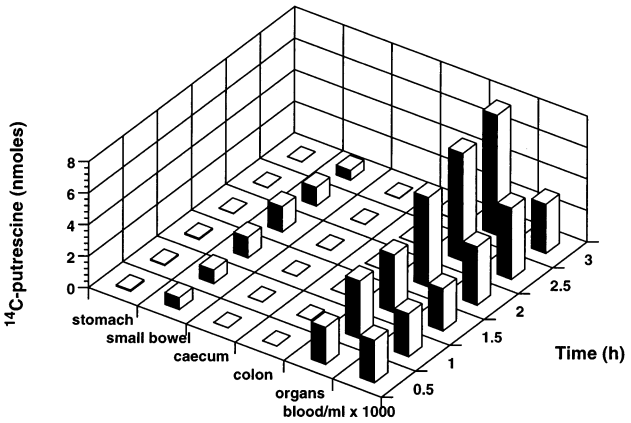

Figure 4 Accumulation of ^{14}C-label (nmoles) in the blood (mL × 1000), stomach, small bowel, cecum, colon, and organs (liver, kidneys, pancreas, spleen, lungs, and carcass) at different times (hours) after intragastric intubation of 8.5 nmoles of ^{14}C-putrescine. Data represent means of five rats. Experimental error was approximately ± 5%.

tric intubation, but increased thereafter. Based on these data, the 2 hour time point was selected to study putrescine uptake/absorption at four different concentrations.

Effect of dose on the uptake of putrescine by the small bowel and body

Absorption of putrescine was determined both indirectly (as above) and directly from the sum of actual radioactivity measured in the organs and carcass (direct method). Putrescine was taken up from the gut lumen by the small intestinal tissue, passed into systemic circulation, and distributed between the internal organs in proportion to the amounts intubated (*Table 1*). The absorption appeared to be linear over the 1000-fold dose range (*Figure 6*). However, the distribution of putrescine was uneven, because the uptake by individual organs differed significantly (as dose/g; *Table 2*). Uptake was proportional to input in the small and large bowels, skeletal muscle, liver, and kidney,

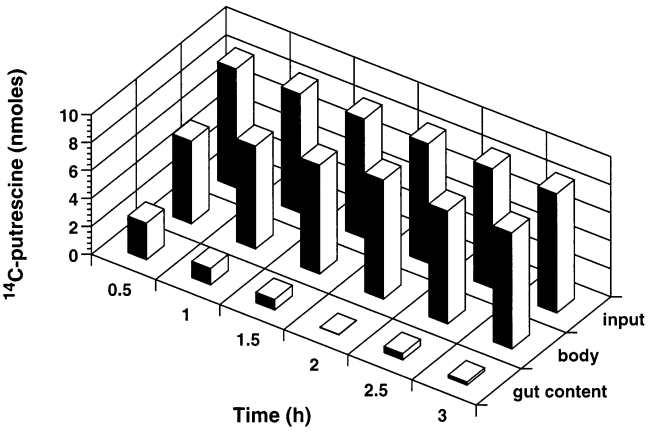

Figure 5 Distribution of ^{14}C-putrescine (8.5 nmoles) between the gut content and body at different times after intragastric intubation of 8.5 nmoles of ^{14}C-putrescine. Data represent means of five rats. Experimental error was approximately ± 5%.

but not in the pancreas and carcass (skin and bone). As putrescine input increased, the total recovery of radioactivity decreased.

Putrescine metabolism

In order to monitor the metabolism of ^{14}C-putrescine, the distribution of ^{14}C-label between polyamines, their acetyl derivatives, and amino acids has been determined by HPLC in samples of plasma, small bowel, liver, and the hind leg gastrocnemius muscle from rats dosed 2 hours previously with 267 nmoles of putrescine. Between 8% and 30% of the ^{14}C-label was associated with polyamines and acetyl polyamines, but most of the label was present in amino acids, indicating that most of the putrescine was oxidized (*Table 3*). Only approximately 10% to 12% of the dose remained as putrescine in the different organs.

Discussion

Putrescine is found in all cells of mammals and, together with the polyamines spermidine and spermine, of which it is a precursor, plays a regulatory role in cellular metabolism.[1–5,34,35] Both infusion of putrescine into the ileal lumen[27] and increase in tissue putrescine content can stimulate mucosal growth, indicating the importance of endogenous putrescine and polyamine sources for the mammalian gut. Endogenous polyamine sources for the gastrointestinal tract include the diet,[10,15,36] bacterial flora,[7,8] enterohepatic circulation,[16] and polyamines released by epithelial cells shed and extruded to the gut lumen. However, the relative contribution of these sources differs in the small and large bowel.

Although the jejunal and duodenal chyme have a high polyamine content, they do not contain any of the prokaryotic or eukaryotic enzymes required for polyamine synthesis.[3,6,8,14,35] Therefore, endogenous contribution of polyamines to small intestinal tissue is needed, the most likely source of which is the diet. (In the healthy jejunum bacterial numbers are low and hence the contribution by bacteria is negligible; data on enterohepatic circulation are contradictory.) In contrast to the small bowel, the decarboxylating activity[16] of the colonic chyme is high. The lumen of the large bowel contains much more putrescine than spermidine or spermine,[16,37] all of which are thought to be produced by the resident bacteria.[34,35] Some of these, such as *Escherichia coli*, have a putrescine exporter protein[38] that facilitates excretion of putrescine synthesized by the bacteria. Most bacteria can synthesize spermidine, but it is not known if they can also excrete it. Only a few microorganisms synthesize spermine.

In the present work we measured the uptake, distribution, and fate of dietary putrescine in healthy rats, because both plant- and meat-based diets are very rich sources of putrescine. In plant food putrescine is present both in free (acid extractable) and conjugated forms (putrescine-derived alkaloids, such as nicotine, scopolamine, and other compounds). In some parts of the plant, conjugated putrescine content can be six times higher than that of free putrescine but it is not known whether putrescine conjugates are degraded by enzymes in the gastrointestinal tract and are available for

Table 1 Tissue accumulation of putrescine 2 hours after intragastric intubation with different doses[1]

	Putrescine input (nmoles)			
	26.7	267	2670	26,700
Stomach	0.07 ± 0.01	0.6 ± 0.2	8.0 ± 0.8	144.6 ± 53.4
Small intestine	3.3 ± 0.6	22.6 ± 1.7	334.4 ± 111	2737 ± 572
Large intestine	0.16 ± 0.04	1.2 ± 0.2	10.5 ± 2.4	235.6 ± 76.4
Liver	0.8 ± 0.4	11.7 ± 3.8	125.3 ± 30.7	1131 ± 84.9
Pancreas	0.04 ± 0.02	0.2 ± 0.02	2.1 ± 0.4	17.7 ± 4.6
Kidney	0.09 ± 0.06	1.1 ± 0.7	10.0 ± 1.1	108.2 ± 26.9
Spleen	0.01 ± 0.002	0.06 ± 0.01	0.4 ± 0.1	5.0 ± 1.6
Lungs	0	0	0.02 ± 0.01	0.4 ± 0.2
Blood[2]	0.1 ± 0.05	0.2 ± 0.03	1.2 ± 0.2	11.5 ± 1.9
Skeletal muscle[3]	1.8 ± 0.6	22.0 ± 7.7	180.9 ± 11.3	1608 ± 473
Carcass	7.1 ± 0.07	44.7 ± 4.6	438.6 ± 110	982.1 ± 81.6
Lumen	6.9 ± 1.1	35.9 ± 5.8	289.8 ± 40.4	4604 ± 1701
Recovery[4] (%)	69.3 ± 3.6	44.4 ± 2.3	45.8 ± 3.5	37.5 ± 3.6

Values represent mean ± SD of five rats.
[1]As calculated from counts originating from ^{14}C-putrescine.
[2]Assuming blood volume is 6 mL.
[3]Assuming skeletal muscle accounts for 40% of total body weight.
[4]Excluding urinary loss and exhaled air.

uptake. If conjugated putrescine is degraded and absorbed, its fate would be the same as that of free putrescine. However, if putrescine conjugates are absorbed intact, we cannot predict their fate.

Dietary putrescine moves along the lumen of the small bowel in a wave-like fashion and is readily absorbed by the gut tissue, as reported by Dorhout and co-workers.[39] The movement of putrescine in the gut lumen was independent of that of proteins and lipids (unpublished data from this laboratory). By 30 minutes, significant amounts of putrescine had already been distributed in the body via the systemic circulation. The highest putrescine absorption was observed at 2 hours, and this time point was selected to study the relationship between the dose of putrescine and absorption/uptake. The dose range chosen was from one-tenth the amounts of putrescine naturally occurring in food in free or conjugated forms to amounts that could never occur naturally.

Based on both direct measurements, which gave the minimal value for putrescine absorption by the gut, and

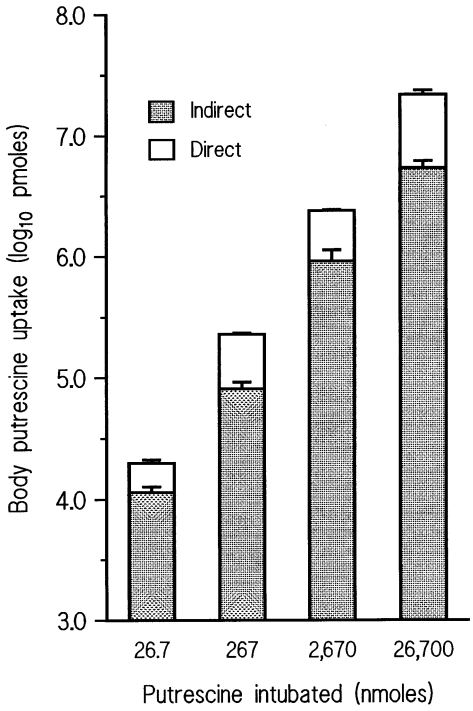

Figure 6 Whole body uptake (log) of dietary putrescine as the function of (log) putrescine input (as pmoles) 2 hours after intragastric intubation with 26.7, 267, 2670, or 26,700 nmoles of putrescine. Indirect uptake by the body was estimated as the difference between input and gut content, whereas direct intake was calculated as the sum of radioactivity measured in organs and carcass. Data represent means ± SE of five rats.

Table 2 Putrescine accumulated in 1 g individual tissues 2 hours after intragastric intubation with different putrescine doses (percent radioactivity/g tissue)[1]

	Putrescine input (nmoles)			
	26.7	267	2670	26,700
Stomach	0.36	0.31	0.43	0.75
Small intestine	3.12	2.18	3.15	2.55
Large intestine	0.59	0.45	0.43	0.77
Liver	0.77	1.13	1.26	1.15
Pancreas	0.29	0.17	0.15	0.13
Kidney	0.31	0.40	0.35	0.41
Spleen	0.14	0.07	0.08	0.08
Lungs	0	0	7.15×10^{-4}	1.5×10^{-3}
Blood[2]	0.03	0.004	0.004	0.003
Skeletal muscle[3]	0.17	0.21	0.17	0.15
Carcass	35.4	22.2	20.7	5.3

Values represent mean of five rats.
[1]As calculated from counts originating from ^{14}C-putrescine.
[2]Assuming blood volume is 6 mL.
[3]Assuming skeletal muscle accounts for 40% of total body weight.

Table 3 Recovery of radioactivity in some organs 2 hours after intragastric intubation of 267 nmoles labeled putrescine

	Distribution of recovered label (%)			
	Serum	Small intestine	Liver	Gastrocnemius muscle
Putrescine	14 ± 3	12 ± 2	10 ± 3	13 ± 2
Spermidine	6 ± 2	2 ± 0	8 ± 0	23 ± 5
Spermine	7 ± 2	4 ± 0	3 ± 1	5 ± 2
Acetyl polyamines	23 ± 4	11 ± 2	5 ± 3	9 ± 2
Amino acids	29 ± 12	69 ± 11	62 ± 12	27 ± 11
Others	21 ± 5	2 ± 0	12 ± 2	23 ± 5

Values represent mean ± SD of five rats.

indirect estimations, which indicated maximum absorption over the entire range of intakes, putrescine appeared to be taken up by the gut tissue by passive diffusion. However, the action of an active transporter that acts as a mop-up mechanism when luminal putrescine concentrations are very low, much lower even than our lowest input of 26.7 nmoles, cannot be excluded. It appeared that the gut could absorb and hold large amounts of putrescine, which was likely to be metabolized or partially oxidized. The release of putrescine from the gut tissue to systemic circulation might also occur by passive diffusion, although it is possible that putrescine metabolites were released and reached the internal organs via the systemic circulation. Putrescine absorption/uptake differed between individual organs and appeared to be related to metabolic activity/requirements (*Table 2*). Unfortunately, such in vivo experiments cannot differentiate between transcellular or paracellular uptake or provide information on the requirements for Na^+ or other ions.

Because putrescine is easily metabolized, acetylated, and oxidized,[24,34] it was important to determine the form in which the ^{14}C-label was present in the samples. Generally, approximately 10% of the label remained in putrescine form, indicating that 10% of dietary putrescine reaches the putrescine body pool. In metabolically active organs, such as the small bowel and liver, 60% to 70% of the putrescine was oxidized, most likely by DAO, and converted to GABA and succinate and then to amino acids via the Krebs cycle. By entering the Krebs cycle, pyruvate reappears as amino acids or as carbon dioxide, which is then exhaled.[40] Combined with losses through urinary excretion, this might explain the poor (38–70%) recovery of radioactivity. In the gastrocnemius muscle, typical of skeletal muscle, less putrescine was oxidized but more was converted to spermidine. In the plasma, 27% of the radioactivity was associated with polyamines, approximately 23% with acetyl polyamines, and approximately 30% with amino acids.

Another interesting observation was that the radiolabel was present in the luminal contents of the colon and cecum within 30 minutes, although the intubated polyamines did not appear to reach the last gut section (i_{17}) by this time (*Figure 2*). A possible explanation for this might be that, as soon as putrescine is taken up by the gut and reaches the systemic circulation, it is distributed in the body. The gut tissue has a polyamine uptake/transport system located on the basolateral side/membrane of the enterocytes that takes up polyamines from the circulation.[20,41] Thus, putrescine can occur in the lumen of the colon before the gavaged putrescine reaches this section of the gut.

In vitro measurements of transepithelial fluxes of putrescine differ in the different gut compartments. Putrescine flux is negative in the ileum, but positive in the jejunum, the normal site of nutrient absorption. All dietary putrescine was fully absorbed from the gut in humans.[42,43]

In summary, dietary putrescine can be readily taken up by the gut, passed to the systemic circulation, and distributed between the internal organs according to their metabolic activity. Some of the putrescine is probably oxidized by DAO by the time it passes through the gut mucosa and reaches the plasma, where a further proportion is oxidized by serum amine oxidases. It appears that approximately 10% of dietary putrescine can contribute to the putrescine pool in the body.

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References

1 Jänne, J., Pösö, H., and Raina, A. (1978). Polyamines in rapid growth and cancer. *Biochim. Biophys. Acta* **473,** 241–293
2 Pegg, A.E. (1986). Recent advances in the biochemistry of polyamines in eukaryotes. *Biochem. J.* **234,** 249–262
3 Tabor, C.W. and Tabor, H. (1984). Polyamines. *Annu. Rev. Biochem.* **53,** 747–790
4 Marton, L. and Morris, D. (1987). Molecular and cellular functions of the polyamines. In: *Inhibition of Polyamine Metabolism* (P.P. McCann, A.E. Pegg, and A. Sjoerdsma, eds.) pp. 79–105. Academic Press, San Diego, USA
5 Heby, O. and Persson, L. (1990). Molecular genetics of polyamine synthesis in eukaryotic cells. *TIBS* **15,** 153–158
6 Bardocz, S., Brown, D.S., Grant, G., and Pusztai, A. (1990). Luminal and basolateral polyamine uptake by rat small intestine stimulated to grow by *Phaseolus vulgaris* lectin phytohaemagglutinin in vivo. *Biochim. Biophys. Acta* **1034,** 46–52
7 Osborne, D.L. and Seidel, E.R. (1989). Microflora-derived polyamines modulate obstruction-induced colonic mucosal hypertrophy. *Am. J. Physiol.* **256,** G1049–G1057
8 Sarhan, S., Knödgen, B., and Seiler, N. (1989). The gastrointestinal tract as polyamine source for tumor growth. *Anticancer Res.* **9,** 215–224
9 Moulinoux, J.-P., Darcel, F., Quemener, V., Havouis, R., and Seiler, N. (1991). Inhibition of the growth of U-251 human glioblastoma in nude mice by polyamine deprivation. *Anticancer Res.* **11,** 175–180
10 Bardocz, S., Grant, G., Brown, D.S., Ralph, A., and Pusztai, A. (1993). Polyamines in food — implications for growth and health. *J. Nutr. Biochem.* **4,** 66–71
11 McCormack, A. and Johnson, L.R. (1991). Role of polyamines in gastrointestinal mucosal growth. *Am. J. Physiol.* **260,** G974–G980
12 Johnson, L.R. (1988). Regulation of gastrointestinal mucosal growth. *Physiol. Rev.* **68,** 456–502
13 Wang, J.-Y. and Johnson, L.R. (1990). Luminal polyamines stimulate repair of gastric mucosal stress ulcers. *Am. J. Physiol.* **259,** G584–G592
14 Wang, J.-Y., McCormack, A., Viar, M.J., and Johnson, L.R. (1991). Stimulation of proximal small intestinal mucosal growth by luminal polyamines. *Am. J. Physiol.* **261,** G504–G511
15 Bardócz, S., Duguid, T.J., Brown, D.S., Grant, G., Pusztai, A.,

White, A., and Ralph, A. (1995). The importance of dietary polyamines in cell regeneration and growth. *Br. J. Nutr.* **73,** 819–828
16 Osborne, D.L. and Seidel, E.R. (1990). Gastrointestinal luminal polyamines: Cellular accumulation and enterohepatic circulation. *Am. J. Physiol.* **258,** G576–G584
17 Bardocz, S. (1993). The role of dietary polyamines. *Eur. J. Clin. Nutr.* **47,** 683–690
18 Seiler, N. (1987). Functions of polyamine acetylation. *Can. J. Physiol. Pharmacol.* **65,** 2024–2035
19 Seiler, N., Bolkenius, F.N., Knödgen, B., and Mammont, P. (1980). Polyamine oxidase in rat tissues. *Biochim. Biophys. Acta.* **615,** 480–488
20 Kumagai, J. and Johnson, L.R. (1988). Characteristics of putrescine uptake in isolated rat enterocytes. *Am. J. Physiol.* **254,** G81–G86
21 Osborne, D.L. and Seidel, E.R. (1988). Putrescine: Rat duodenal lumen concentrations and transport by duodenal crypt cells in culture. *FASEB J.* **2,** A735
22 Groblewski, G.E., Hargittai, P.T., and Seidel, E.R. (1992). Ca^{2+}/calmodulin regulation of putrescine uptake in cultured gastrointestinal epithelial cells. *Am. J. Physiol.* **262,** C1356–C1363
23 Seidel, E.R., Haddox, M.K., and Johnson, L.R. (1985). Ileal mucosal growth during intraluminal infusion of ethylamine or putrescine. *Am. J. Physiol.* **249,** G434–G438
24 Sessa, A. and Perin, A. (1994). Diamine oxidase in relation to diamine and polyamine metabolism. *Agents Actions* **43,** 69–77
25 Raul, F., Gosse, F., Galluser, M., Hasselmann, M., and Seiler, N. (1995). Functional and metabolic changes in intestinal mucosa of rats after enteral administration of ornithine α-ketoglutarate salt. *J. Parent. Ent. Nutr.* **19,** 145–150
26 Bardócz, S., Grant, G., Brown, D.S., and Pusztai, A. (1998). Putrescine as a source of instant energy in the small intestine of the rat. *Gut* **42,** 24–28
27 Seidel, E.R., Haddox, M.K., and Johnson, L.R. (1984). Polyamines in the response to intestinal obstruction. *Am. J. Physiol.* **246,** G649–G653
28 Ginty, D.D., Osborne, D.L., and Seidel, E.R. (1989). Putrescine stimulates DNA synthesis in intestinal epithelial cells. *Am. J. Physiol.* **257,** G145–G150
29 Luk, G.D. and Baylin, S.B. (1983). Polyamines and intestinal growth — increased polyamine biosynthesis after jejumectomy. *Am. J. Physiol.* **245,** G656–660
30 Hosomi, M., Stace, N.H., Lirussi, F., Smith, S.M., Murphy, G.M., and Dowling, R.H. (1987). Role of polyamines in intestinal adaptation in the rat. *Eur. J. Clin. Invest.* **17,** 375–385
31 Grant, G., Dorward, P.M., and Pusztai, A. (1993). Pancreatic enlargement is evident in rats fed diets containing raw soybeans (*Glycine max*) or cowpeas (*Vigna unguiculata*) for 800 days but not in those fed diets based on kidney beans (*Phaseolus vulgaris*) or lupinseed (*Lupinus angustifolius*). *J. Nutr.* **123,** 2207–2215
32 Lowry, O.H., Rosebrough, N.J., Farr, A.L., and Randall, R.J. (1951). Protein measurements with the Folin phenol reagent. *J. Biol. Chem.* **193,** 265–275
33 Seiler, N. and Knödgen, B. (1980). High-performance liquid chromatography procedure for the simultaneous determination of natural polyamines and their monoacetyl derivatives. *J. Chromatogr.* **221,** 227–235
34 Seiler, N., Delcros, J.G., and Moulinoux, J.-P. (1996). Polyamine transport in mammalian cells. An update. *Int. J. Biochem. Cell Biol.* **28,** 843–861
35 Seidel, E.R. and Scemama, J.-L. (1997). Gastrointestinal polyamines and regulation of mucosal growth and function. *J. Nutr. Biochem.* **8,** 104–111
36 Okamoto, A., Sugi, E., Koizmi, Y., Yanagida, F., and Udaka, S. (1997). Polyamine content of ordinary foodstuffs and various fermented foods. *Biosci. Biotech. Biochem.* **61,** 1582–1584
37 Hinuma, K., Maghsoudloo, M., Murphy, G.M., and Dowling, R.H. (1992). Dietary and intestinal polyamines in the rat: in vitro transport studies. In: *Polyamines in the Gastrointestinal Tract* (R.H. Dowling, U.R. Fölsch, and C. Löser, eds.) pp. 463–472. Kluwer Academic Press, Dordrecht, The Netherlands
38 Kashiwagi, K., Miyamato, S., Suzuki, F., Kobayashi, H., and Igarashi, K. (1992). Excretion of putrescine by the putrescine-ornithine antiporter encoded by the potE gene of *Escherichia coli*. *Proc. Natl. Acad. Sci. USA* **89,** 4529–4533
39 Dorhout, B., van Fassen, A., van Beusekom, C.M., Kingma, A.W., Dehoog, E., Nagel, G.T., Karrenbeld, A., Boersma, E.R., and Muskiet, F.A.J. (1997). Oral administration of deuterium-labelled polyamines to sucking rat pups: Luminal uptake, metabolic fate and effects of gastrointestinal maturation. *Br. J. Nutr.* **78,** 639–654
40 Seiler, N., Bolkenius, F.N., and Rennert, O.M. (1981). Interconversion, catabolism and elimination of the polyamines. *Med. Biol.* **59,** 334–346
41 Dumontier, A.M., Brachet, P., and Tomé, D. (1992). Putrescine transport in the rabbit small intestine mounted in the Ussing chamber. In: *Polyamines in the Gastrointestinal Tract* (R.H. Dowling, U.R. Fölsch, and C. Löser, eds.) pp. 453–461. Kluwer Academic Publishers, Dordrecht, The Netherlands
42 Milovic, V., Odera, G., Murphy, G.M., and Dowling, R.H. (1997). Jejunal putrescine absorption and the "pharmacokinetics"/biotransformation of ingested putrescine, in humans. *Gut* **41 (Suppl 3),** A62
43 Benamouzig, R., Mahe, S., Luengo, C., Rautureau, J., and Tomé, D. (1997). Fasting and post-prandial polyamine concentrations in the human digestive lumen. *Am. J. Clin. Nutr.* **65,** 766–770