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Research Paper

Brain uptake and distribution patterns of 2-hydroxypropyl-ß-cyclodextrin after intrathecal and intranasal administration

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Abstract

Objectives Cyclodextrins are increasingly used therapeutically. For example, 2-hydroxypropyl-ß-cyclodextrin (kleptose) is used for the treatment of Niemann–Pick disease. Kleptose crosses the blood–brain barrier poorly, in part because of a central nervous system (CNS)-to-blood (efflux) transporter, and so is administered by the intrathecal (IT) route in the treatment of Niemann–Pick disease.

Methods Here, we evaluated the uptake and distribution of kleptose by the brain and spinal cord after intranasal (IN) or IT delivery.

Key findings Kleptose distributed to all regions of the brain and spinal cord after IN administration, with only 3.27% of the administered dose entering the bloodstream. Intrathecal delivery produced levels in the whole brain about 40 times higher than intranasal administration and about 20 times higher than previously found after intravenous administration. About 70–90% of the IT dose rapidly entered the bloodstream, in part because of the previously described efflux transporter. The uptake by CNS after IN and IT was both non-saturable. The uptake by the olfactory bulb, hypothalamus and pons-medulla was favoured by all routes of administration.

Conclusions Kleptose was taken up by all regions of the CNS after either IN or IT administration, but IN administration resulted in only a fraction of the uptake of the IT route.

Keywords: cyclodextrin; blood–brain barrier; intrathecal; intranasal; central

Introduction

Cyclodextrins, a class of cyclized oligosaccharides originally isolated from bacterial digests of plant starches, are finding an increasing number of uses in pharmacology and pharmaceutics.[1, 2] Their cone shape has a hydrophilic outer surface and a hydrophobic inner surface, allowing them to interact with hydrophobic molecules or regions of molecules, rendering them more water soluble and chemically stable. Widely used as excipients, they improve the solubility of hydrophobic molecules.

Cyclodextrins are categorized as α, β *or* γ cyclodextrins based upon whether their rings contain 6, 7 or 8 glucopyranoside units, respectively.[1] Molecular weights range between 972 and 1297 Da, and toxicity ranges from 1 to 4 g/kg in mice. These basic structures can be modified in various ways by the addition of functional groups, which modify their properties. For example, 2-hydroxypropyl-ß-cyclodextrin (kleptose) is used as an excipient for a variety of drugs administered via the rectal, buccal, ophthalmic, oral and intravenous routes.[1, 3]

One approach combines cyclodextrins with peptides, which alters their regional brain distribution after intranasal administration.[4–8] Other work reported that combining peptides with cyclodextrins alters the distribution pattern in the brain of those peptides.[9, 10] Furthermore, the distribution pattern is different depending on which cyclodextrin is used.

Perhaps most exciting is the use of cyclodextrins as single agents for their therapeutic value. Currently, kleptose is used in the treatment of Niemann–Pick disease,[11, 12] an autosomal recessive lysosomal storage disease resulting in cholesterol and glycosphingolipid build up in peripheral tissues and brain cells. Kleptose given peripherally in mice decreased intraneuronal storage of glycosphingolipid and cholesterol from neurons, decreased neuroinflammation, decreased neurodegeneration, and significantly prolonged life.[12–14] In humans, however, the peripheral administration of kleptose resulted in less dramatic improvements in the brain function and so is often given by the intrathecal route.[15, 16]

Understanding the pharmacokinetics and brain penetration characteristics of cyclodextrins would aid in deciding questions such as the routes of administration and doses needed to be used to produce effects in the central nervous

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system (CNS). Previous work has shown that radioactive kleptose crosses the blood–brain barrier (BBB) by a non-saturable mechanism.[17] However, the rate of brain uptake (0.235 µl/g-min) and the percent of the administered dose entering the brain (0.03 %Inj/g) are low and are not detectable at all in the spinal cord. One reason for this poor uptake is the presence of a brain-to-blood efflux system that seems to be particularly robust in the spinal cord. Herein, we investigate the potential of intranasal and intrathecal kleptose as an alternative to intravenous delivery to the CNS.

Materials and Methods

Studies reported here followed a preset design. The decision to collapse data over time rather than analyse as time-series data was made *post hoc* when preliminary analysis showed a lack of significant correlation of dependent values with time.

Radioactive materials

The cyclodextrin kleptose [(2-hydroxy-[2-^{14}C]propyl)-β-cyclodextrin] was radioactively labelled with ^{14}C (C-Klep) by the Institute of Isotopes Co. Ltd (Budapest, Hungry). The specific activity was 0.251 MBq/mg. The ^{14}C samples were processed with Solvable (PerkinElmer) and incubated overnight at 68°C until completely solubilized. Ecoscint (National Diagnostics) was then added, and their levels of radioactivity were measured in a beta counter (TriCarb 3110TR; PerkinElmer). Bovine serum albumin (Sigma-Aldrich, St. Louis, MO) was labelled with ^{125}I with the chloramine-T method (I-Alb) and purified on a column of Sephadex G-10.

Intranasal delivery

CD-1 male mice, 8 to 10 weeks old, purchased from Charles River Laboratories (Wilmington, MA) were housed on a 12/12 hour light/dark cycle with ad libitum food and water at the Veterans Affairs Puget Sound Health Care System, an American Association for Accreditation of Laboratory Animal Care (AAALAC)-approved facility. Protocols were approved by the Veterans Affairs – Puget Sound Institutional Animal Care and Use Committee. Mice were anaesthetized to effect with about 4 mg/kg of intraperitoneal urethane (40% solution). C-Klep (10^6 dpm/mouse) in 1 µl of phosphate-buffered saline was injected intranasally near the cribriform plate by inserting a gel-loading pipette 4 mm into the left nostril of the mouse. At each of various times after intranasal delivery (2.5, 5, 10, 20, 30, 45, 60, 90 and 120 min), 2 to 3 mice had their right carotid artery severed, arterial blood collected and were then decapitated. The brain was removed and dissected (frontal cortex, parietal cortex, occipital cortex, hippocampus, hypothalamus, thalamus, striatum, cerebellum, midbrain, pons-medulla and olfactory bulb) as was the spinal cord (cervical, thoracic and lumbar), and all regions were weighed. Levels of radioactivity were determined, and results were expressed as the percent of the injected material per gram of tissue (%Inj/g):

$$\%\text{Inj/g} = 100\,(\text{R})\,/(\text{wI})$$

where R is the dpm for a brain region, w is the weight of the brain region and I is the amount of radioactivity administered by the intranasal route. The percent of the injected material present in arterial serum (%Inj/ml) was calculated by the following equation:

$$\%\text{Inj/ml} = 100\,(\text{S})\,/(\text{vI})$$

where S is the dpm in the arterial serum sample and v is the volume of the arterial serum sample.

Other mice were treated as above except that some had 0.15 mg of unlabelled kleptose included in the intranasal injection. These mice were studied 20 min after the injection, and selected regions (olfactory bulb, frontal cortex, hypothalamus, cerebellum, lumbar spine and arterial serum) were evaluated.

Intrathecal delivery

CD-1 male mice, 8 to 10 weeks old, purchased from Charles River Laboratories (Wilmington, MA) were housed on a 12/12 hour light/dark cycle with ad libitum food and water at the Veterans Affairs Puget Sound Health Care System, an AAALAC-approved facility. Protocols were approved on 7 March 2018, by the local Institutional Animal Care and Use Committee (#0933). Mice were anaesthetized to effect with about 4 mg/ml of intraperitoneal urethane (40% solution). Erioglaucine disodium salt (Brilliant Blue FCF; Sigma-Aldrich, St. Louis, MO) was injected into other mice, and the whole brain and spinal cord were harvested 10 min later. In other mice, C-Klep (10^6 dpm/mouse) in 10 µl of phosphate-buffered saline was injected into the intrathecal space at the intersection of the dorsal midline and gluteus medius just superior to the lumbosacral junction. At each of various times after the injection (2.5, 5, 10, 20, 30, 45, 60, 90 and 120 min), 2 to 3 mice had their right carotid artery severed, arterial blood collected and were then decapitated. The brain was removed and dissected (frontal cortex, parietal cortex, occipital cortex, hippocampus, hypothalamus, thalamus, striatum, cerebellum, midbrain, pons-medulla and olfactory bulb) as was the spinal cord (cervical, thoracic and lumbar), and all regions were weighed. Levels of radioactivity were determined, and results were expressed as the percent of the injected material per gram of tissue (%Inj/g) and %Inj/ml using the equations above.

Other mice were given an intrathecal injection of 1.5(10^6) counts per minute (CPM) of albumin radioactively labelled with ^{125}I. Carotid artery blood was collected 10 min after the injection, and the %Inj/ml was calculated as above.

Other mice were treated as above except that they had 1.5 mg of unlabelled kleptose included in the intranasal injection. These mice were studied 20 min after the injection, and selected regions (olfactory bulb, frontal cortex, hypothalamus, cerebellum, lumbar spine and arterial serum) were evaluated.

Statistics

Calculations and comparisons of regression lines for relations between %Inj/g versus time or %Inj/ml versus time were performed using Prism 8.0 (GraphPad Inc, San Diego, CA), and $P < 0.05$ taken to indicate a significant relation between %Inj/g or %Inj/ml and time. Means for time series data were calculated by collapsing data across time and are expressed with their standard errors. Mice receiving unlabelled kleptose were compared with those only receiving C-Klep by analysis

of variance (ANOVA) followed by Sidak's multiple range test. The area under the curve (AUC) for arterial serum was calculated using Prism 8.0 for 0 to 30 min.

Results

Distribution after intranasal administration

Radioactivity was detected in all samples after the intranasal administration of C-Klep but varied as a function of time for only arterial serum, thoracic spine and striatum. Temporal patterns for serum and striatum and two regions whose radioactive levels did not correlate with time are shown in Figure 1. The serum AUC was 16.1 (%Inj/ml)-min, which gives a bioavailability of 3.27% based on the AUC data for intravenous injection previously published.[13] This small amount of material entering the blood, the general lack of correlations with tissue uptake over time and the low rate of uptake for the thoracic spine and striatum argue that blood-to-brain entry was not a major factor in the uptake of material into the CNS after intranasal administration.

Figure 2 shows the distribution of C-Klep by the brain region after intranasal administration. The olfactory bulb showed the highest uptake with little variation among the other regions. The inclusion of unlabelled kleptose in the C-Klep injection did not alter the uptake values for any of the regions tested (data not shown). Therefore, the nose-to-brain route appears to be non-saturable for the uptake of kleptose.

Distribution after intrathecal administration

Figure 3 shows the distribution of dye after its intrathecal administration. Dye is most intense in the lumbar region, which is the region of injection, and least intense in the cervical spine region. No dye was seen in the brain (not shown).

All regions showed radioactivity after the intrathecal administration. All regions showed a correlation with time regarding the uptake except for the lumbar, thoracic and cervical spinal regions and the occipital and parietal cortices (Table 1). Figure 4 shows examples of the temporal effect on %Inj/g. All regions showing a correlation between %Inj/g and time had values that increased over time except for arterial serum, which decreased over time. The serum AUC was 360 (%Inj/ml)-min, which gives a bioavailability of 73% based on the AUC data for intravenous injection previously published.[13] This shows that the majority of material injected by the intrathecal route rapidly entered the bloodstream.

To determine whether the IT injection method was inadvertently injecting material directly into the bloodstream, I-Alb was injected IT and arterial blood samples collected 10 min later. The brain/serum ratio for I-Alb 10 min after its IT injection was 24.9 +/– 10.6 (%Inj/ml), which is about half of the expected brain/serum value after intravenous injection of I-Alb.

Figure 5 shows the values of %Inj/g collapsed over time for the various brain regions after IT injection. Consistent with the findings with dye, the lumbar spine values are very high with a rapid decrease in thoracic and cervical spines. The

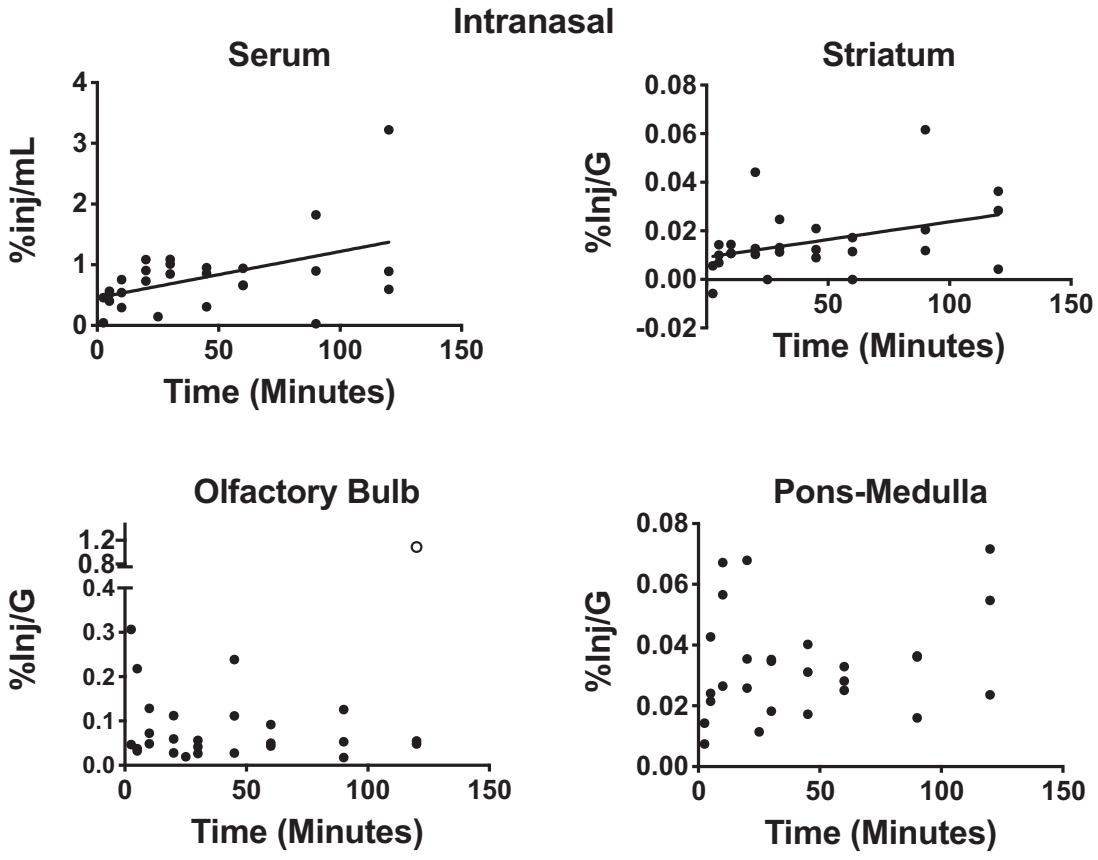

Figure 1 Kinetics of radioactive kleptose after its intranasal delivery. The upper left panel shows the slow entry into the bloodstream of a small amount of the administered dose. The upper right panel shows a slow increase over time of radioactivity into the striatum. Lower panels show non-time-dependent uptake of radioactivity by the olfactory bulb (open circle is an outlier excluded from regression analysis) and pons-medulla.

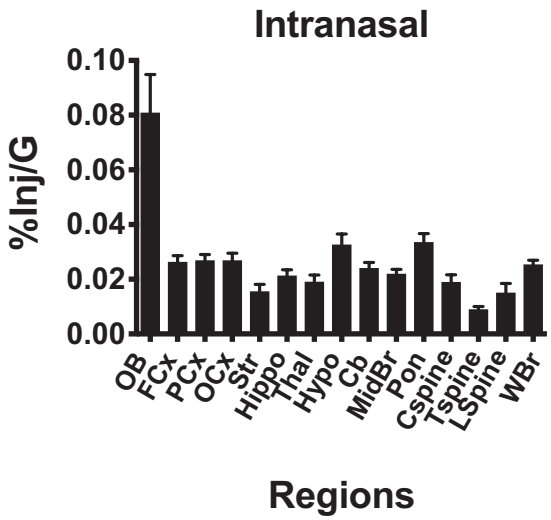

Figure 2 Distribution pattern of radioactive kleptose after its intranasal administration for the brain regions, whole brain and spinal cord regions. The highest levels in the brain are for the olfactory bulb, hypothalamus and pons-medulla. Abbreviations: OB, olfactory bulb; FCx, frontal cortex; PCx, parietal cortex; OCx, occipital cortex; Str, striatum; Hippo, hippocampus; Thal, thalamus; Hypo, hypothalamus; Cb, cerebellum; MidBr, midbrain, Pon, pons-medulla; Cspine, cervical spinal cord; Tspine, thoracic spinal cord; Lspine, lumbar spinal cord; WBr, whole brain.

olfactory bulb and the hypothalamus show the next highest levels, although they are more remote from the spinal cord. Of the regions closest to the cervical spine, the pons-medulla but not the midbrain or cerebellum also showed a higher level of uptake. This suggested that the uptake by brain regions was a function of affinity for C-Klep.

The inclusion of unlabelled kleptose in the IT injection had no statistically significant effect on the uptake.

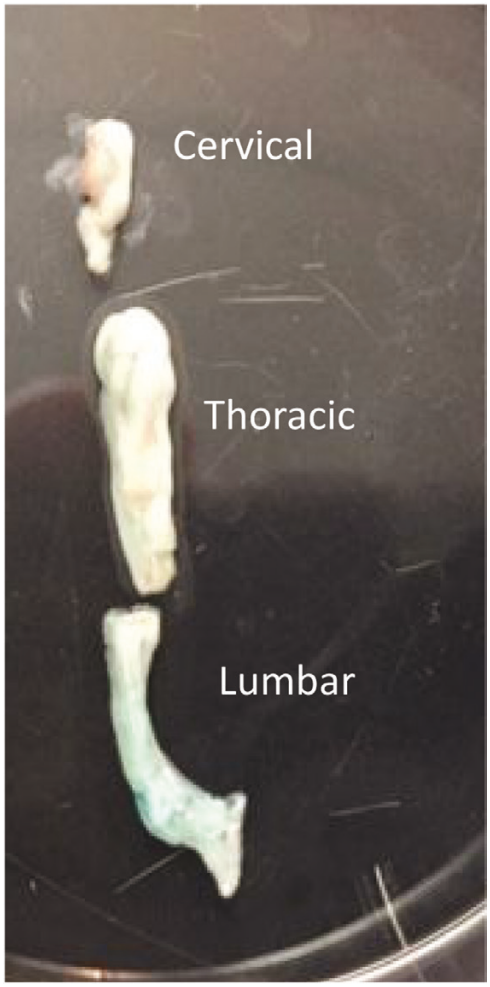

Figure 3 Distribution of erioglaucine disodium salt after its intrathecal injection. Dye could be seen in the spinal cord with lumbar > thoracic > cervical. No dye could be seen in the brain (not shown).

Discussion

These studies compared the IN and IT routes of administration in their abilities to deliver the cyclodextrin kleptose to the brain. Previously, we and others have published regarding the BBB penetration and peripheral pharmacokinetics for kleptose after intravenous administration.[13, 17] Here, we examined the brain uptake, distribution and related questions for kleptose after its intranasal or intravenous administration.

The intranasal administration to the level of the cribriform plate has emerged as a way to deliver a number of substances to the brain.[7, 8, 18–22] Here, we found that kleptose was taken up immediately by all brain regions, although at a low percentage of the injected dose. A very small amount, estimated to be 3.27%, of the intranasal dose of kleptose entered the bloodstream. Levels increased with time for blood, striatum and thoracic spine, but for other regions, levels of kleptose quickly reached a steady state. This ability for a substance to quickly distribute to all regions of the brain is a characteristic of the intranasal route previously observed,[10, 18, 22] but the mechanism for this is unclear. The lack of further increase and the small amount of kleptose moving in the brain-to-blood direction suggests that kleptose is quickly sequestered and retained by brain tissue.

With IT injection, both dye and radioactivity showed a decreasing level of uptake as one ascended the spinal cord, with the uptake by the C-spine being much lower than uptake by the L-spine. No dye was seen in the pons-medulla, the brain region adjacent to the C-spine, and radioactive levels were about 1/10th of those of the C-spine, showing a sharp drop off as kleptose entered the cranium. Unlike after IN administration, the level of kleptose gradually increased in most brain regions with time after IT administration. Spinal cord levels remained unchanged, however. Given that the CSF volume is about 10% of brain weight (see page 22 in Bradbury's textbook[23]) and the mouse spinal cords weighed about 100 mg, the CSF space of these mice can be estimated to be about 10 µl , and so it is likely that the 10 µlvolume of the IT injection displaced much of the spinal cord CSF, allowing sequestration of high levels of kleptose that did not subsequently change with time. The unsequestered amount of kleptose would be free to enter the cranial space along with the general flow in the CSF from the spinal cord towards the sites of CSF reabsorption, the arachnoid villi and the extracranial lymphatics,[24, 25] resulting in the observed increase in the brain levels over time.

An unusual feature of the IT administration is the very high levels of kleptose appearing immediately in the blood. The pattern was similar to that of IV injection, with the highest levels being at the earliest time points and suggested that about 73% of the material injected via IT administration immediately

Table 1 %Inj/g versus time (min) after intrathecal injection

Region	Intrathecal injection			
	m[1]	i[1]	r^2	*n* (*P*<)
Olfactory bulb	.052 +/– 0.021	0.85 +/– 1.12	0.200	27 (0.05)
Frontal cortex	0.013 +/– 0.0046	0.061 +/– 0.26	0.256	27 (0.01)
Parietal cortex			0.05	27 (NS)
Occipital cortex	0.0072 +/– 0.0035	0.31 +/– 0.020	0.143	27 (*P* = 0.052)
Striatum	0.0081 +/– 0.0033	0.30 +/– 0.19	0.191	27 (0.05)
Thalamus	0.013 +/– 0.0046	0.081 +/– 0.26	0.248	27 (0.01)
Hypothalamus	0.067 +/– 0.029	0.644 +/– 1.6	0.180	27 (0.05)
Hippocampus	0.0095 +/– 0.0036	0.22 +/– 0.21	0.213	27 (0.05)
Cerebellum	0.0092 +/– 0.0042	0.24 +/– 0.24	0.161	27 (0.05)
Midbrain	0.022 +/– 0.0071	(–)0.045 +/– 0.41	0.275	27 (0.01)
Pons-medulla	0.065 +/– 0.028	0.58 +/– 1.6	0.175	27 (0.05)
Whole brain	0.018 +/– 0.0071	0.22 +/– 0.41	0.207	27 (0.05)
Cervical spinal cord			0.09	27 (NS)
Thoracic spinal cord			0.04	27 (NS)
Lumbar spinal cord			0.01	27 (NS)

[1]m is the slope of the linear relation in units of %Inj/(g-min) and i the Y-intercept of the linear relation in units of %Inj/g. The r^2 relates to the goodness of fit of the line and *n* is the number of animals constituting the linearity. (*P*<) is level of statistical significance with NS being Not Significant.

entered the blood. Two explanations could underlie this pattern: (1) much of the IT administration was injected into the vertebral vessels and so directly entered the bloodstream and (2) kleptose was transported rapidly in the brain-to-blood direction by the previously described brain efflux system.[13] The latter is primarily located in the distal spinal cord. Interestingly, five outliers shown in Figure 4 as open circles had much lower levels in the blood than the other data points. These points also tended to have the highest values in spinal cord regions (but less so for brain regions). Including these five points indicated that 73% of the IT kleptose entered the blood, but this value was above 90% if they were excluded. To distinguish between the possibilities of direct injection into the bloodstream versus transporter, we injected radioactive albumin, a molecule that typically only slowly enters the bloodstream as CSF is reabsorbed, using the same method as that for IT kleptose and dye. We found that about half of the IT injection of radioactive albumin appeared in the blood. This substantial level suggests that about half of the IT dose is directly injected into the bloodstream, probably by way of the vertebral vessels. However, this amount is still less than 70–90% and so likely kleptose is also being transported out of the spinal cord and into the blood by the previously described transporter. A previous study found, in the cynomolgus monkey, that about 40% of the IT injection appeared in blood by 5 h.[26] This could be attributed to CSF reabsorption from the CNS to the blood, but it is interesting that the values for %Inj/g for the heart after IT injection were closer to those found after IV injection than after injection into the lateral ventricle of the brain. The latter observation could be explained by the CNS-to-blood transporter located at the spinal cord.

The transporter responsible for kleptose transport is unknown. Previous work has shown that excess unlabelled kleptose enhances, rather than inhibits, its rate of transport, suggesting stochastic kinetics.[13] This study shows that, on the one hand, the efflux rate is very high in the spinal cord, but, on the other hand, the lack of change in the %Inj/g for the spinal cord regions suggests that kleptose is quickly incorporated into tissue and then no longer available for efflux. The CNS has many efflux systems located at the vascular BBB and the choroid plexus.[27] Of these transporters, P-glycoprotein (P-gp) has a very diverse number of ligands.[28] However, cyclodextrins inhibit P-gp activity, possibly through modulation of the membrane microenvironment[29] and so would not be expected to be transported by P-gp. Another possibility is that the transporter does not recognize kleptose but rather has an endogenous ligand that binds kleptose so that both are transported out of the CNS. Inhibition of this transporter should greatly increase the amount of kleptose accumulating in the brain after IT administration. However, its presence means that IT administration is also delivering kleptose to peripheral tissues, possibly in doses that can treat peripheral disease as well.

Kleptose was taken up by all brain and spinal cord regions studied, regardless of whether the route of administration was IT, IV or IN. However, the percent uptake differed tremendously for these routes. Comparing values for the whole brain, the %Inj/g was about 0.06 after IV, about 0.025 after IN and almost 1% after IT. This means that the amount of drug taken up was nearly 40 times greater after IT than after IN, despite the finding that 70–90% of the IT dose entered the blood almost immediately after administration. Therefore, little of the drug found in the brain after IT injection made its way there by first entering the blood and then crossing the BBB. These findings have a practical application to the current administration of kleptose in clinical conditions.[11] While IT dosing has inherent risks associated with it, it is currently the only route that can adequately deliver therapeutically relevant CNS doses. Both IV and IN would require 20- and 40-fold higher doses to reach therapeutic levels in the brain, which could be limited by off-target toxicities.

An unusual finding is that three regions of the brain had an especial predilection for the uptake of kleptose, no matter

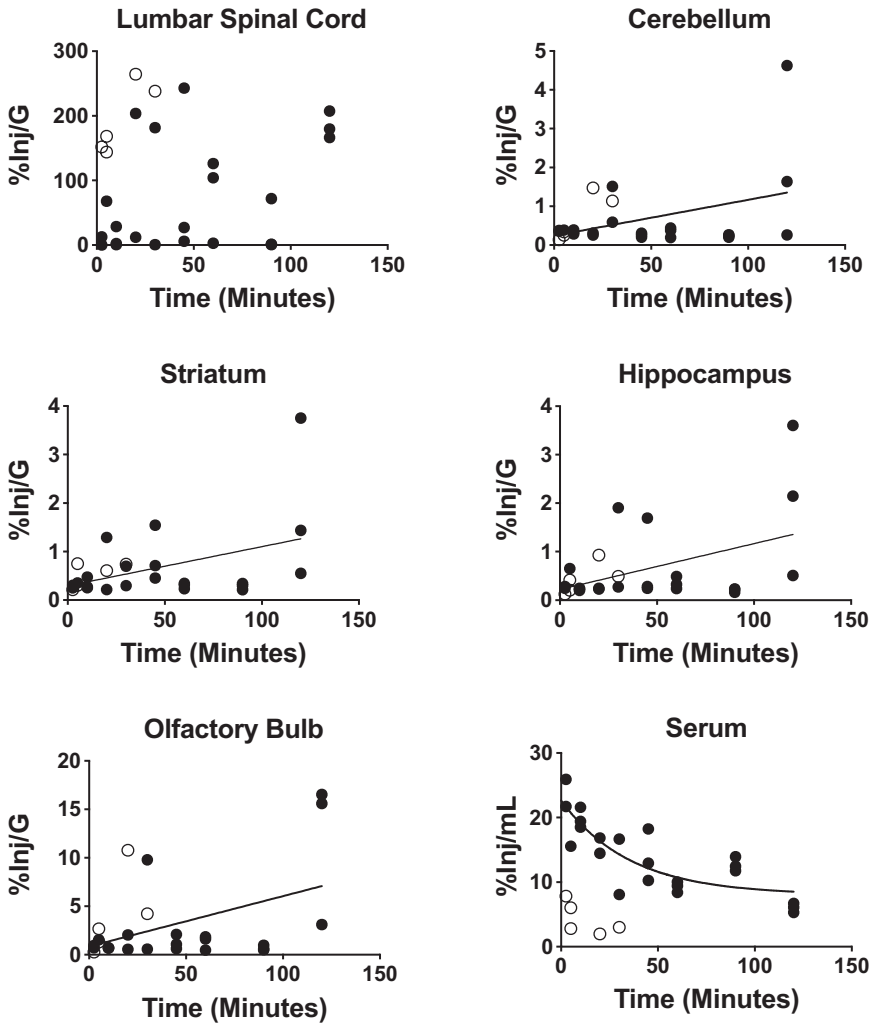

Figure 4 Kinetics of radioactive kleptose after its intrathecal delivery. The upper left panel shows non-time-dependent radioactive levels in the lumbar spine. The lower right panel shows time-dependent clearance of radioactivity from the blood; the curve resembles the kinetics of intravenous injection. The remaining panels show representative time-dependent increases in the brain regions. The open circles represent animals that had a much lower %Inj/ml than the other animals.

what the route of administration. The olfactory bulb, pons-medulla and hypothalamus all had higher uptakes than any other brain region regardless of the route of administration. IN delivery would be thought to favour the uptake by the olfactory bulb as that region is the closest to the site of administration, whereas IT would disfavour it as in that case it is furthest from the site of administration. By similar reasoning, the uptake by the pons-medulla would be favoured by the IT administration and disfavoured by IN administration. Hypothalamus is not obviously favoured by either route of injection. Nevertheless, after all three routes of administration, the highest levels of uptake were the olfactory bulb, hypothalamus and pons-medulla. Why kleptose favours these regions is unknown.

An other important finding is that we could not find evidence for saturability of kleptose uptake after either route of administration. This is an important finding for clinical administration in that it suggests that the amount of kleptose reaching a target site can be proportionately increased by increasing the amount of kleptose administered.

These results are not likely to have resulted from the degradation of kleptose to a radioactive fragment. Labelling was with a ^{14}C that was an integral part of the structure of kleptose, and so a degradation product could have only occurred with degradation of kleptose itself. Kleptose, like most cyclodextrins, is not metabolized after parenteral administration but secreted intact in urine.[30]

In conclusion, we found that kleptose is taken up by all regions of the brain and spinal cord after either IT or IN administration. The uptake, however, is about 40 times greater after IT administration, making this a more likely therapeutic route for CNS delivery compared with nasal or IV administration. CNS tissue uptake by these routes appeared to be non-saturable, and distribution and sequestration by the brain and spinal cord tissues are rapid by either route of administration. Interestingly, the pons-medulla, hypothalamus and olfactory bulb have a particular avidity for kleptose regardless of route of administration. The therapeutic value of these observations is yet unknown and warrants further study.

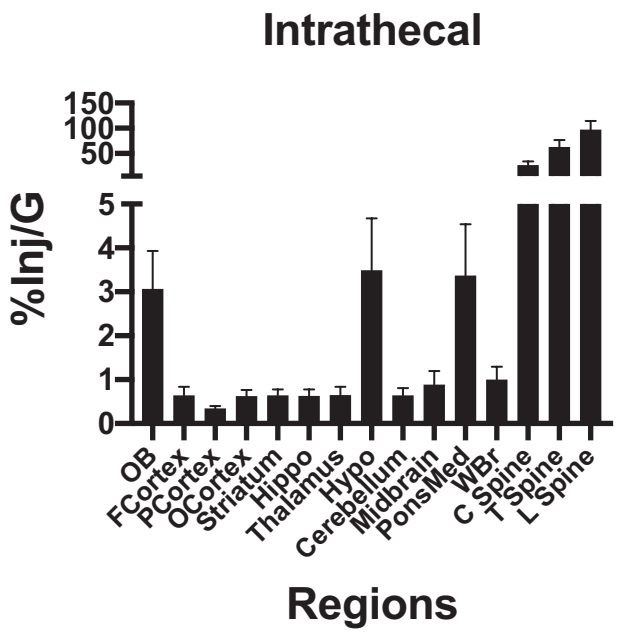

Figure 5 Distribution pattern of radioactive kleptose after its intrathecal administration for brain regions, whole brain and spinal cord regions. Note highest levels in the brain are for the olfactory bulb, hypothalamus and pons-medulla. Abbreviations: OB, olfactory bulb; FCortex, frontal cortex; PCortex, parietal cortex; OCortex, occipital cortex; Hippo, hippocampus; Hypo, hypothalamus; PonsMed, pons-medulla; WBr, whole brain; C Spine, cervical spinal cord; T Spine, thoracic spinal cord; L Spine, lumbar spinal cord.

Author Contributions

All the authors participated in research design and wrote or contributed to the writing of the manuscript; K.M.H. and K.M.B. conducted the experiments; P.C. and K.E. contributed to reagents and analytic tools; W.A.B. performed the data analysis.

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Conflicts of Interest

The authors have no competing interests.

Data availability

Data available from authors upon request.

References

1. Davis ME, Brewster ME. Cyclodextrin-based pharmaceutics: past, present, and future. *Nat Rev Drug Discov* 2004; 3: 1023–35. http://doi.org/10.1038/nrd1576
2. Varca GHC, Andreo-Fiho N, Lopes PS et al. Cyclodextrins: an overview of the complexation of pharmaceutical proteins. *Curr Protein Pept Sci* 2010; 11: 255–63. https://doi.org/10.2174/138920310791233387
3. Loftsson T, Brewster ME. Pharmaceutical applications of cyclodextrins: basic science and product development. *J Pharm Pharmacol* 2010; 62: 1607–21. https://doi.org/10.1021/js950534b
4. Davis SS, Illum L. Absorption enhancers for nasal drug delivery. *Clin Pharmacokinet* 2003; 42: 1107–28. http://doi.org/10.2165/00003088-200342130-00003
5. Merkus FW, Verhoef JC, Marttin E et al. Cyclodextrins in nasal drug delivery. *Adv Drug Deliv Rev* 1999; 36: 41–57. http://doi.org/10.1016/s0169-409x(98)00054-4
6. Irie T, Uekama K. Cyclodextrins in peptide and protein delivery. *Adv Drug Deliv Rev* 1999; 36: 101–23. http://doi.org/10.1016/s0169-409x(98)00057-x
7. During MJ, Cao L, Zuzga DS et al. Glucagon-like peptide-1 receptor is involved in learning and neuroprotection. *Nat Med* 2003; 9: 1173–9. https://doi.org/10.1038/nm919
8. Frey WH II. Bypassing the blood-brain barrier to deliver therapeutic agents to the brain and spinal cord. *Drug Deliv Technol* 2002; 2: 46–9.
9. Nonaka N, Farr SA, Kageyama H et al. Delivery of galanin-like peptide to the brain: targeting with intranasal delivery and cyclodextrins. *J Pharmacol Exp Ther* 2008; 325: 513–9. https://doi.org/10.1124/jpet.107.132381
10. Nonaka N, Farr SA, Nakamachi T et al. Intranasal administration of PACAP: uptake by brain and regional brain targeting with cyclodextrins. *Peptides* 2012; 36: 168–75. http://doi.org/10.1016/j.peptides.2012.05.021
11. Calias P. 2-Hydroxypropyl-Beta-cyclodextrins and the blood-brain barrier: considerations for Niemann-Pick disease type C1. *Curr Pharm Des* 2017; 23: 6231–8. http://doi.org/10.2174/1381612823666171019164220
12. Davidson CD, Nafeeza FA, Matthew C et al. Chronic cyclodextrin treatment of murine Niemann-Pick C disease ameliorates neuronal cholesterol and glycosphingolipid storage and disease progression. *PLoS One* 2009; 4: e6951. http://doi.org/10.1371/journal.pone.0006951
13. Banks WA, Engelke K, Hansen KM et al. Modest blood-brain barrier permeability of the cyclodextrin kleptose: modification by efflux and luminal surface binding. *J Pharmacol Exp Ther* 2019; 371: 121–9. http://doi.org/10.1124/jpet.119.260497
14. Liu B, Turley SD, Burn DK et al. Reversal of defective lysosomal transport in NPS disease ameliorates liver dysfunction and neurodegeneration in the npc1$^{-/-}$ mouse. *Proc Natl Acad Sci USA* 2009; 17: 2377–82. http://doi.org/10.1073/pnas.0810895106
15. Matsuo M, Shraishi K, Wada K et al. Effects of intracerebroventricular administration of 2-hydroxypropyl-beta-cyclodextrin in a patient with Niemann-Pick type C disease. *Mol Genet Metab Rep* 2014; 17: 391–400. http://doi.org/10.1016/j.ymgmr.2014.08.004
16. Matsuo M, Togawa M, Hirabaru K et al. Effects of cyclodextrin in two patients with Niemann-Pick type C disease. *Mol Genet Metab* 2013; 108: 76–81. http://doi.org/10.1016/j.ymgme.2012.11.005
17. Pontikis CC, Davidson CD, Walkley SU et al. Cyclodextrin alleviates neuronal storage of cholesterol in Niemann-Pick C disease without evidence of detectable blood-brain barrier permeability. *J Inherit Metab Dis* 2013; 36: 491–8. http://doi.org/10.1007/s10545-012-9583-x
18. Banks WA, During MJ, Niehoff ML. Brain uptake of glucagon-like peptide-1 antagonist exendin(9-39) after intranasal administration. *J Pharmacol Exp Ther* 2004; 309: 469–75. http://doi.org/10.1124/jpet.103.063222
19. Benedict C, Hallschmid M, Schmitz K et al. Intranasal insulin improves memory in humans. *Psychoneuroendocrinology* 2004; 29: 1326–34. http://doi.org/10.1016/j.psyneuen.2004.04.003
20. Kastin AJ, Pan W. Intranasal leptin: blood-brain barrier bypass (BBBB) for obesity? *Endocrinology* 2006; 147: 2086–7. http://doi.org/10.1210/en.2006-0208
21. Lochhead JJ, Thorne RG. Intranasal delivery of biologics to the central nervous system. *Adv Drug Deliv Rev* 2012; 64: 614–28. http://doi.org/10.1016/j.addr.2011.11.002
22. Salameh TS, Bullock KM, Hujoel IA et al. Central nervous system delivery of intranasal insulin: mechanisms of uptake and effects on cognition. *J Alzheimers Dis* 2015; 47: 715–28. http://doi.org/10.3233/JAD-150307

23. Bradbury M. *The Concept of a Blood-Brain Barrier*. London: John Wiley and Sons Ltd, 1979.
24. Boulton M, Flessner M, Armstrong D et al. Contribution of extracranial lymphatics and arachnoid villi to the clearance of a CSF tracer in the rat. *Am J Physiol* 1999; 276: R818–23. http://doi.org/10.1152/ajpregu.1999.276.3.R818
25. Davson H, Segal MB. *Physiology of the CSF and Blood-Brain Barriers*. London: CRC Press, 1996, 489–523.
26. Calias P, Papisov M, Pan J et al. CNS penetration of intrathecal-lumbar idursulfase in the monkey, dog and mouse: implications for neurological outcomes of lysosomal storage disorder. *PLoS One* 2012; 7: e30341. http://doi.org/10.1371/journal.pone.0030341
27. Taylor EM. The impact of efflux transporters in the brain on the development of drugs for CNS disorders. *Clin Pharmacokinet* 2002; 41: 81–92. http://doi.org/10.2165/00003088-200241020-00001
28. Begley DJ. ABC transporters and the blood-brain barrier. *Curr Pharm Des* 2004; 10: 1295–312. http://doi.org/10.2174/1381612043384844
29. Fenyvesi F, Fenyvesi E, Szente L et al. P-glycoprotein inhibition by membrane cholesterol modulation. *Eur J Pharm Sci* 2008; 34: 236–42. http://doi.org/10.1016/j.ejps.2008.04.005
30. Loftsson T, Moya-Ortega MD, Alvarez-Lorenzo C et al. Pharmacokinetics of cyclodextrins and drugs after oral and parenteral administration of drug/cyclodextrin complexes. *J Pharm Pharmacol* 2016; 68: 544–55. http://doi.org/10.1111/jphp.12427