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Development of recto-dispersible tablets and granulate capsules for the treatment of serious neonatal sepsis in developing countries

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Abstract

Current paediatric antibiotic therapies often use oral and parenteral routes of administration. Neither are suitable for treating very sick neonates who cannot take oral medication and may be several hours away from hospital in developing countries. Here, we report on the development of rectal forms of ceftriaxone, a third-generation cephalosporin, along with an absorption enhancer, sodium chenodeoxycholate. Recto-dispersible tablets and capsules were developed and successfully passed 6-month accelerated stability tests. Rabbit bioavailability showed plasma concentrations above the minimal inhibitory concentrations for 3 formulations of recto-dispersible tablets and 2 formulations of hard capsules. Clinical batches are currently being prepared for human evaluation with the prospect of offering therapeutic alternatives for treating critically ill neonates. This proof of concept for efficient rectal delivery of antibiotics could help the development of other rectal antibiotic treatments and increase options for non-invasive drug development for paediatric patients.

Main text

Introduction

Neonatal sepsis is an invasive bacterial infection. Neonates in developing countries are at particular risk of death with around 1 million deaths occurring within 24 hours of birth and another million within the first week of life ¹. Although the leading causes of death are pre-term births, events related to delivery, sepsis, meningitis and pneumonia together account for 21% of all newborn deaths ². Neonates that die in the first few days of life suffer from diseases and conditions that are associated with poor quality of care and reduced access to effective treatment ². Most deaths occur in or near home. Substantial reductions in deaths from neonatal infections can be achieved with prompt administration of effective injectable antibiotics ^{3–5}. However, simpler ways of administering antibiotic treatment reliably at home have recently been tested for children with no signs of severe sepsis ^{6,7}. These are more likely to succeed by reducing the number of injections, increasing access and adherence to treatment but their results cannot be extended to babies excluded from evaluation – low-birth weight infants or babies with signs of critical illness such as unconsciousness and convulsions; these children are at highest risk of

death when the disease progresses rapidly. Evidence from previous trials indicates that between 10-76% of parents refuse referral in severe infections ⁸⁻¹¹. For most antibiotics, oral and injectable treatments are available, but neither can be used in the vicinity of patients who are too ill to take oral therapy and cannot reach hospital for several hours.

Ceftriaxone (CTX) is a semi-synthetic broad-spectrum third-generation cephalosporin antibiotic ¹². Its antibacterial spectrum covers both Gram-positive and Gram-negative bacteria. It has good Central Nervous System penetration ¹³ and a good safety record ¹⁴ which makes it suitable for antibiotic treatment of possible severe neonatal sepsis. The drug binds strongly to human serum proteins and hence the elimination half-life of CTX is age-dependent (15 to 19 hours in 1-8-day neonates compared to 8h in adults), which favors its development and use as a single daily dose in critically ill neonates. CTX excretion is through urine and bile (40-60% and 10-20% respectively) and does not change with dose ^{15,16}. The clinical efficacy of CTX is dependent upon the time during which free drug concentrations in serum remain above minimum inhibitory concentration (MIC, reportedly at or below 1µg/ml for most sensitive bacteria ^{17,18}) and concentrations above the MIC are required for more than 50% of the dosing interval to be effective ¹⁹.

CTX is highly hydrophilic and hydro-soluble, with a low octanol/water partition coefficient ($\log P = -2.1 \pm 0.2$) ²⁰ and is thus poorly absorbed through mucosal membranes and categorized as class 3 in the Biopharmaceutics Classification System (BCS) ²¹.

CTX has only been registered for use as an extemporaneously reconstituted injectable solution. It is sensitive to degradation in gastric fluid and is poorly absorbed from the intestinal tract ¹², which favors rectal compared to oral administration. However, CTX must be combined with an absorption enhancer to achieve adequate rectal absorption. A human bile salt, sodium chenodeoxycholate (NaCDC) in a ratio 1/4 with CTX, has previously been shown as the optimal absorption enhancer for CTX ²². Pre-formulation studies of CTX have been performed ²³ and these confirmed poor compatibility with most of liquid excipients but good compatibility with NaCDC, thus supporting the development of solid drug dosage formulations ²³.

Previous studies have suggested that rectal CTX therapy can achieve clinically relevant plasma concentrations in animals and man, and hence CTX was developed into rectal formulations that could be administered in the community to a neonate with a rapidly evolving serious sepsis infection, while awaiting transfer to hospital ²². The formulations developed and tested by Hoffmann La Roche (Roche), the originator of CTX, was a wax suppository, which was not practical as a dosage form for tropical rural settings with high humidity and high day-time temperatures. Liquid dosage forms have been previously described, but these did not comply with standard requirements for further development ²⁴.

The aim of this study was to develop a stable and bioavailable solid CTX-NaCDC drug dosage form (recto-dispersible tablet or capsule), and compare bioavailability with the rectal wax suppository developed and tested by Roche. Solid dose medications currently on the market are tablets and capsules. Neither are commonly used for rectal administration, but some data on rectal use can be found in the literature. Rectal hard capsules are occasionally used in hospitals for pediatric patients when adjustment of the dose to the bodyweight is necessary but no commercialized form is yet available ²⁵. Recto-dispersible tablets currently do not exist on the market, but a chewable lamotrigine dispersible tablet has shown significant absorption when administered rectally ²⁶. Therefore a first step was to develop the rationale for recto-dispersible mucoadhesive tablets based on the available literature and then develop rectal solid dosage forms of CTX with NaCDC as an absorption enhancer. Finally, *in vitro* and *in vivo* evaluation of the formulations are described. Success in this specific application of rectal antibiotic therapy as an alternative to oral or injectable treatments could have relevance for other antibiotics and medications for patients who can neither take oral medications nor access injectable treatment.

Materials and Methods

Materials

The active pharmaceutical ingredient used was Ceftriaxone hemiheptahydrate sodium salt from Discovery Fine Chemicals (batch n° 73777). The excipients used were of pharmaceutical grade, as follows: Sodium chenodeoxycholate (NaCDC) from Biosynth, Switzerland (batch n° 0000011434); Emcompress (DP, Dicalcium phosphate) from Laboratoire Mendell, France (batch 88661/0); Pearlitol 400 and 100SD (Mannitol) from Roquette, France; Corn starch (CS) from Cooper, France; Lactose monohydrate 35µm and 75µm from Cooper, France; Hydroxypropylmethylcellulose E 50LV (HPMC)

from Colorcon, USA; Polyvinylpyrrolidone (PVP K30) from Cooper, France; Maltodextrin from Cooper, France; Crospovidone (Kollidon CL, BASF, France); Croscarmellose sodium from Cooper, France ; Anhydrous colloidal silica (Aerosil 300, Cooper France); Magnesium stearate from Cooper, France; Talc from Cooper, France; Pregelatinized corn starch (starch derivative, Sepistab ST200) from Seppic, France; Microcrystalline cellulose (MCC, Avicel PH102) from Fagron, France; Magnesium Aluminometasilicate (Neusilin) from Fuji Chemical Industry, France.

The solvents used for analyses were of analytical grade. Water was deionized by osmosis, using “Système special dialyse Fernand Lopez, Toulouse, France” equipped with MILLERHOF 42250434 cartige.

Methods

Granulation

Where applicable, all powder blends (e.g. CTX and NaCDC +/- diluent, binder) were mixed in the Turbula powder mixer (10min, 34 rpm, Turbula T2F, France) prior to granulation. For wet granulation in a fluidized bed granulator (Miniglatt®, Glatt Ingenieurtechnik GmbH, Germany) with top down nozzle, the input air temperature was set at 70°C, water spray pressure at 0.9 bar with a water flow rate around 4g/min. Granules were dried at 70°C until complete dryness, with a powder temperature not exceeding 45°C. For 130g of powder, optimal conditions required 33g of water and 6min of drying, followed by 6min of cooling before opening the apparatus.

When granulated “traditionally”, the powder mixture of CTX, NaCDC and PVP K30 was moistened with water in a planetary mixer (Kitchenaid®, France) and passed through a sieve (mesh 355µm) for granulation. The granules were dried in an oven (Mettmert UNE 200-800, Germany) for 24 hours at 40°C.

The moisture content was measured using loss on drying (LOD) technique on 5g of powder at 150°C for 15min (Mettler Toledo analyzer MJ33, France). The results were expressed as weight percentage. Granules with a LOD around 7% were considered as dried, as some CTX levels of moisture (3.5 hydrate equivalent to approximately 9% w/w) had been shown to be more labile ²³.

Alternatively, dry granulation was performed using a tablet press (Korsh Pressen Germany, n°K0000114, 2002) with 14mm flat round punches for CTX compaction. The tablets obtained were

crushed with small mortar and pestle then passed through a sieve of 1.25mm. Granules could be further separated using sieves of 200 and 800 μ m.

Preparation of CTX recto-dispersible tablets

The intra-granular phase was mixed with the excipients of the extra-granular phase using a Turbula powder mixer (10min, 34 rpm). This extra-granular phase included the bioadhesive (HPMC), the super-disintegrant (crospovidone), a flowability improver (silica/talc) and the lubricants (magnesium stearate and talc). The flowability of the final mixture was then characterized before being compressed according to European Pharmacopoeia standards.

The compression was performed on a laboratory scale tablet press (Korsh Pressen Germany, No. K0000114, 2002) using a set of flat round punches (diameter 14mm).

Tablets were sealed in alu/alu sealed blisters and stored at 4°C and evaluated for hardness, disintegration, friability and dissolution according to European Pharmacopoeia standards and analyzed using an HPLC method.

Preparation of CTX capsules

The CTX-NaCDC powder blend or the fluid bed granulated CTX-NaCDC granules were filled into hard shell capsules. The Capsules were sealed in alu/alu blisters and stored at 4°C for further analyses.

Preparation of drug dosage forms for an animal study

Tablets were prepared as described in the paragraph above. For the rabbit studies, the tablet mass was adjusted for rabbit weight by cutting the tablet to obtain one entire piece that was placed into a 1mL syringe to maintain the tablets' integrity during storage and transport. Syringes containing tablets adjusted to each rabbits' bodyweight were sealed into alu/alu blisters and kept in a fridge until use.

Capsules n°4 were manually filled with CTX and NaCDC powders or CTX-NaCDC granules to adjust the weight of their content to the bodyweight of each rabbit. They were stored in alu/alu blisters at 4°C and administered using a home-made blunt cut-edge syringe.

Suppositories were prepared with 20% w/w CTX, 5% NaCDC and 75% Suppocire AML, as formulated by Roche and tested in humans ²². Powders were dispersed using a magnetic stirrer at 40°C in melted Suppocire.

Samples to be used in testing in rabbits were molded in a pre-weighted 1mL syringe with a cut end. After solidification, the quantity was individually adjusted to each rabbit's bodyweight by cutting the excess weight of the suppository. Samples were then sealed in alu/alu blisters and kept in the fridge until use. All formulations were tested for batch homogeneity and content using HPLC method before use.

Bulk and tapped density

The bulk densities and tapped densities of the mixtures and granules were evaluated using a measuring cylinder placed on a tapping machine (Vankel Tap density, Varian, France) that subjected the mixture to a specified number of taps (10, 250, etc). The different densities were obtained by direct reading of the volume of the mixture in the measuring cylinder after weighing a certain quantity of powder.

Flowability

The flowability of powders, mixtures and grains was evaluated through a 10mm funnel in accordance to the flow test of the European pharmacopoeia (European Pharmacopoeia 9) using a PharmaTest PTG-S4 (PharmaTest, France) apparatus. An exactly weighed quantity of powder was introduced in the funnel and the time of flow across the orifice was measured. The results are a mean of three replicates.

Hardness

Tablet hardness was measured using a European Pharmacopoeia hardness tester (PTB 311E, PharmaTest, France).

Disintegration

Tablet disintegration was measured according to the European Pharmacopoeia method using an Erweka ZT501 (France) apparatus. The tablet disintegration time was noted after their complete disintegration in 1L distilled water at 37°C.

Dissolution

Dissolution was measured using a paddle apparatus (AT7, Sotax, France), filled with 900mL 50mM phosphate buffer at pH 7.5 and at 37°C. The stirring rate was set at 50 rpm. Samples were taken after

filtration with 10µm prefilters at defined times and analyzed using the UV absorbance at 270nm. The content was derived from a 2-40µg/mL linear standard curve ($R^2>0.998$). Preliminary studies (not detailed here) showed the UV spectroscopic method was linear, reproducible and that up until 14h no significant degradation of CTX had occurred (content was checked in parallel by the HPLC method).

Stability evaluation

Formulations were sealed after manufacture into Alu/Alu blisters and then tested in a climatic chamber at 40°C/75% relative humidity (RH) (ICH accelerated aging conditions) or at local, uncontrolled atmospheric conditions (mean 21°C, 55-60%RH). The CTX content was analyzed at T0 (initial value at zero time) and after 1 month, 3 months and 6 months of accelerated aging using the HPLC method and compared to the CTX content obtained at T0. The results were expressed as % of the initial CTX content.

HPLC analysis of CTX formulations

The HPLC assay was carried out using an HPLC system (SpectraSystem, France) with a C18 reverse phase column (YMC ODS-H80 150 x 4.6mm, 4µm), a pump (P1000), an auto-sampler (AS3000), a DAD detector (UV6000), a degasser (Vacuum degasser, Biotech AB, France). The method with minor modifications had previously been validated and described ²⁷. Briefly, the method was based on ion-pairing reverse phase liquid chromatography, the mobile phase was a combination of 18mM tetrabutylammonium bromide diluted in phosphate buffer and methanol (70:30, v/v) and the buffer solution was composed of a mixture of $\text{KH}_2\text{PO}_4/\text{K}_2\text{HPO}_4$ at 25mM, pH 7.5. This buffer was also used for sample preparation and dilutions. The volume of injection was 10µL.

Animal CTX bioavailability assessment and analysis

Bioavailability in rabbits was carried out at the DETERCA animal facility laboratory of the Bordeaux University (animal facility agreement n° A33-0.63-235, animal experiment authorization and ethical committee agreement DIR 13123).

Six white New Zealand healthy male adult rabbits (Eurolap, France) were used per group of rectally delivered CTX solid formulations and compared with the CTX rectal wax suppository control group and the reference group of intravenous CTX. Animals were housed individually and provided with standardized rabbit pellets and water *ad libitum*. Animals were weighed and restrained the day before

the treatment. All formulations were dosed and individually adjusted at 20mg/kg of body weight. Administration was preceded by rectum emptying using an inflatable balloon. The IV control formulation was administered using a catheter in the left ear as a one-minute injection. Blood samples were withdrawn into 4mL lithium heparinized tubes at predefined times (t₀, 5min, 15min, 30min, 45min, 1h, 2h, 4h, 8h and 24h) using a 24G catheter in the right ear. Samples were centrifuged immediately at 2500rpm for 10min. Plasma was frozen immediately at -20°C and plasma concentration was determined by HPLC method.²² Bioavailability was calculated from individual plasma profiles using trapezoidal rule. After the treatment, rabbits were either kept as pets by individual volunteers or by the White Rabbit Association, or sacrificed by a lethal injection of sodium pentobarbital after an injection of a sedative (Calmivet®).

Statistical analysis

Statistical analyses were performed using Excel Student test. Differences were considered significant at a threshold level of $p < 0.05$.

Results and Discussion

Rationale for recto-dispersible tablets

Our target product profile (TPP) was stable solid drug dosage forms, recto-dispersible tablets or capsules, exhibiting adequate bioavailability and short T_{max}, which would enable prompt treatment of probable serious neonatal sepsis in high burden developing countries, where hospital treatment is often refused or difficult. Rectal CTX would facilitate immediate administration and be stable in tropical conditions. A single 500mg dose was evaluated, but further dose to weight adjustments may be required at a later date. A rectal treatment would have all the advantages in terms of stability and ease of administration, but achieve rapid drug release, comparable with traditional fatty suppositories which are not practical because they would melt in tropical conditions.

Our development was based on existing oro-dispersible formulations. To avoid premature expulsion, mucoadhesive agents commonly used in vaginal^{28,29} or sublingual^{30,31} tablets were evaluated. The most commonly used mucoadhesive polymers³² form a hydrophilic matrix and provide sustained release of the active pharmaceutical ingredient (API) which can be counterbalanced with a super-

disintegrant. We examined excipients such as piroxicam^{30,31}, carvediol³³ and fentanyl³⁴ which allow optimal exposure of the API (and bioadhesive retention), dicalcium phosphate and mannitol as fillers (mannitol also has bioadhesive properties with high dissolution), croscopovidone, sodium croscarmellose or starch glycocholate as disintegrants with bioadhesive properties, magnesium stearate (as lubricant) for formulating mucoadhesive super-disintegrant tablets^{30,31,33,34}. These studies concerned BCS 2 class APIs and we found no formulation developed associating an absorption enhancer (commonly considered as necessary for oral administration of BCS 3 API), with both a mucoadhesive and a super-disintegrant. Direct compression of oral or buccal mucoadhesive tablets is reported using carbomer or HPMC as mucoadhesive excipients^{35–37}. The bile salt derivative, sodium glycodeoxycholate used as absorption enhancer did not negatively affect the tablet adhesiveness which made it acceptable for a buccal bioadhesive drug delivery system³⁷.

Pre-formulation studies²³ pointed to some key constraints: poor flowability and moisture lability. Therefore, direct compression or dry granulation, which do not require wetting the powder blend, appeared essential to prevent water induced instability. Our formulation was intended for the rectal route and an important consideration was the limited quantity of fluid available in the rectum for disintegration and dissolution of CTX³⁸. Therefore, hydrophilic and water attracting excipients were of special interest. Furthermore, introducing a solid form in the rectum can activate the defecation reflex in the baby. The size of the solid form therefore needed to be as small as possible.

In view of the required properties for dispersible (oral) tablets and the rectal route for systemic administration, we considered the following tablet compositions as a starting point for further development of recto-dispersible tablets:

- 500mg CTX as the active pharmaceutical ingredient
- 125mg NaCDC as an absorption enhancer, as previously published²²
- A mucoadhesive agent to avoid early leakage from rectum
- A super-disintegrant to help disintegration in the small quantity of liquid available in the rectal lumen

The balance between the mucoadhesive agent and disintegrant would need to allow complete and rapid drug release while conferring some mucoadhesive properties to the disintegrated granules.

- Lubricants and glidants
- Filler

For better local tolerance the total weight of the tablet containing 500mg was limited to 1g.

Design and in vitro evaluation of recto-dispersible tablets and capsules of CTX

Direct compression was evaluated with various proportions of glidants (colloidal silica, talc, starch, or a mixture of them), direct compression fillers (model excipient used was dicalcium phosphate) and a mixture of these excipients. These did not improve flowability of CTX or the CTX-NaCDC mixture which was infinite according to European Pharmacopoeia standards. Dry and wet granulation were evaluated to solve flowability problems provided that drying was carried out rapidly after wetting and that moderate compression forces were used ²³. Wet granulation provided optimal results if precise control and a short CTX wetting time was used; to limit crystal damage a moderate compression using 80MPa obviated the significant PXRD broadening, which was seen with 150MPa compression ²³.

Granulation of CTX alone or a CTX-NaCDC blend was tested using fluid bed technology, traditional wet granulation technology (blender and oscillating granulator) or dry granulation (compaction, crushing, calibration) with PVP K30 as model binder. A limit of 2%w/w of PVP could be used by fluid bed technology for technical reasons. The results are summarized in Table 1.

Dry granulation was a viable alternative to wet granulation provided that moderate compression forces were used. The granule densification was greater compared to fluid bed granulation, but the yield was lower with our equipment (<70% compared to wet granulation with >85%) and the specific dry formulation equipment is required. 5% PVP traditional granulation increased the density of granules compared to fluid bed granulation with 2% PVP, but the flowability was impaired. The flowability and bulk density were comparable in formulations with 2% PVP prepared using fluid bed and traditional granulation respectively. Fluid bed wet granulation was therefore considered as a means to control technical parameters and reduce wetting time of CTX, providing better long-term stability.

For tablet manufacture, lubricants and glidants were used in the usual proportions (0.6% of talc, magnesium stearate and colloidal silica mixture in 1/1/1 ratio) to avoid technical problems (such as tablet capping) during compression even if the preliminary studies on granules did not show any significant

reduction of flowability (data not shown). The required mucoadhesive properties were obtained by using HPMC.

The ratio between disintegrant and mucoadhesive was optimized to obtain a visible gel layer and adherent properties to the disintegration disk whilst showing the fastest disintegration time. A range of HPMC contents from 2 to 12% was evaluated, with disintegrant ranging from 2 to 7 percent. The combination of 2% HPMC and 7% disintegrant gave the fastest disintegration time while still providing adherence properties to the disintegration disks. Binders and fillers were further screened in tablet formulation. Table 2 summarizes the optimization of the extra-granular and the intra-granular phases of recto-dispersible tablets and the results obtained.

HPMC as a mucoadhesive excipient is often considered as mucoadhesive agent of choice^{39,40} and has recently shown higher mucoadhesiveness compared to other common mucoadhesive agents such as guar gum, chitosan or Eudragit® RS⁴¹. A 5% HPMC formulation of sublingual sumatriptan tablets enabled a complete release of API in less than 15 minutes, which was faster than polyox or Carbopol examined as alternative mucoadhesive agents in the same formulation⁴². Disintegration times, obtained using discs in the European Pharmacopoeian disintegration apparatus, were reportedly higher in mucoadhesive formulations due to adhesion of hydrated polymer gel to the disc⁴². This was likely to increase the *in vivo* residence time³⁴. As higher disintegration times made it easier to distinguish between formulations, we used this method to screen formulations.

The drug release for all fillers with tested formulations was very rapid (more than 85% in less than 15 minutes) and thus considered as similar according to current guidelines⁴³. Dissolution was not a discriminant parameter for selecting an optimized formulation, but technical considerations and stability appeared to be of importance.

In view of these first results, PVP K30 was retained as a binder (but free flowing granules could be obtained without binder, cf. Table 1) and calcium phosphate, MCC and mannitol were kept as fillers for further characterization and pre-stability evaluation. Optimization concerned the granulation and compression parameters and when CTX and NaCDC were granulated with water only (without binder), filler was added in the extra-granular phase. The optimum drying conditions were assessed using LOD and was considered complete when 7-8% of water was recovered (corresponding to the part of crystalline water of CTX left after drying²³). Approximately 33g of water was necessary for granulation of 130g of powder blend.

The tablets were compressed using flat round punches, which are suboptimal for friability. Results are considered as preliminary because the shape would need further adaptation for rectal insertion (e.g. oblong) during industrial scale-up. Hardness was easily adjusted.

Dry granulated CTX and NaCDC were formulated into tablets (Table 3) and evaluated. Dicalcium phosphate and mannitol were screened as fillers in comparable formulations to wet granulation tablets. Mannitol in formulations posed difficulties for hardness adjustment. Low tablet hardness caused high friability. Longer disintegration times were obtained for dicalcium phosphate compared to mannitol (6.4 and 3 minutes respectively), as expected from the filler solubility and tablet hardness. Compared to wet granulation results, the disintegration time of mannitol dry granulation tablets was similar (3 minutes for both) and the preliminary stability results for mannitol and dicalcium phosphate were both satisfactory. Friability (including an appropriate tablet shape) needed further optimization.

The preliminary stability of the mannitol tablet with PVP and the dicalcium phosphate tablet with PVP achieved poor stability (preliminary stability <75% after 6 months at 40°C/75%RH) and were therefore discarded. The dicalcium phosphate optimized tablet (wet granulated, without PVP) did not comply with stability requirements and was discarded. The MCC tablet with and without PVP passed stability requirements, but tablet hardness would need to increase to avoid any friability problems. The results are summarized in Table 4.

Given these results, our final choice was fluid bed wet granulated CTX and NaCDC for reasons of higher yield, easier technical feasibility and controlled process parameters with our laboratory equipment. Three formulations of wet granulated recto-dispersible tablets were finally chosen, using CTX and NaCDC granulated using fluidized bed technology and water as the granulation solvent;

- Tablet containing mannitol as a filler, without binder
- Tablet containing MCC as a filler without binder
- Tablet containing MCC as a filler and PVP as a binder.

Capsules (CTX-NaCDC powder-filled and CTX-NaCDC fluid bed granulate-filled) were developed as alternative formulations and their preliminary stability was assessed (Table 4). Due to the poor flowability of CTX powder, powder-filled capsules were not considered as potential alternatives for commercial

manufacture, except as a pharmacokinetic control for other formulations. Gelatin capsules were used or all hard shell capsules, as faster disintegration time is reported compared to hypromellose shells ⁴⁴. Granules filled in capsules were obtained without any binder, using water as the wetting solvent with fluidized bed technology.

In vivo evaluation of CTX recto-dispersible tablets compared to capsules

The 5 chosen formulations, 3 recto-dispersible tablets and 2 capsules, were tested in rabbits for bioavailability against the IV reference and fatty-wax suppository controls. Dosage was individually adjusted to the bodyweight of each rabbit to the equivalent of 20mg/kg CTX and administered rectally. Although the fatty-wax formulation did not comply with our TPP, Roche had previously shown that the product provided adequate bioavailability in humans ²². The pharmacokinetic parameters are summarized in Table 5 and plasma profiles are given in Figure 1 (individual profiles Figure 1A and comparison of mean profiles Figure 1B).

For ethical reasons the number of animals used was limited, resulting in relatively high standard deviations. None of the differences in C_{max}, AUC, time above MIC and absolute or relative bioavailability (F and F' respectively) between the Roche suppository control formulation and new rectal forms were statistically significant (Student test, $p > 0.05$), although the capsule containing powder and tablet (MCC and PVP) appeared to reach higher bioavailability of CTX. The T_{max} was significantly longer for both capsules ($p < 0.02$, Student test), but not for tablets. The MCC containing recto-dispersible tablets were expelled by rabbits in some cases (2/6 and 3/6 for MCC tablet with and without PVP respectively), but this was not observed for the mannitol tablets or for capsules. One suppository (control formulation) was also expelled. This could be related to the very dry conditions of a specific rabbit's rectum and might not occur in humans. The MCC and PVP-containing tablets seemed to have optimal *in vivo* characteristics, but if expulsion is an important risk, the mannitol tablet could represent the best compromise for the formulation of recto-dispersible tablets. MCC and PVP-containing tablets showed a shoulder on the plasma profile, which might be attributed to enterohepatic recycling of CTX ⁴⁵. Interestingly, this shoulder was less visible on the other PK profiles. The bioavailability of powder-

filled and granulate-filled capsules were obtained. Both showed good plasma profiles for CTX, but the capsule filled with powder would not be of value in industrial scale-up because of poor flowability.

Conclusion

In conclusion, three recto-dispersible tablet formulations were successfully designed, characterized and gave good CTX plasma profiles. These results provide proof of concept that recto-dispersible tablets and granulate-filled capsules might be viable alternatives to common rectal drug dosage forms, with the advantages of stability and ease of manufacture. They could be used for noninvasive administration of ceftriaxone in conditions where very sick neonates with probable septicemia cannot access or refuse hospital referral. Alternatively, hard capsules filled with granules might be formulated, but it is likely that this will increase capsule size and reduce bioavailability. Clinical batches of the formulations are currently being produced for human clinical trial evaluation.

Beyond this case example of ceftriaxone formulations, our study provides proof of concept for rectal medicines as an alternative to oral and injectable treatments.

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M. Gomes is a former staff member of the World Health Organization (WHO); the authors alone are responsible for the views expressed in this publication and they do not necessarily represent the decisions, policy or views of the WHO.

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Figures' legends :

Figure 1: (A) Individual rabbit plasma profiles per formulation administered rectally ; (B) Mean rabbit plasma profiles of CTX rectal formulations in rabbits (truncated X axis between 8h and 24h).

Plasma profiles of rabbits after administration of ceftriaxone suppository

Plasma profiles of rabbits after administration of ceftriaxone (CTX+CDC) powder capsules

Plasma profiles of rabbits after administration of ceftriaxone (CTX+CDC) grain capsules

Plasma profiles of rabbits after administration of ceftriaxone Avicel without PVP tablets

Plasma profiles of rabbits after administration of ceftriaxone Mannitol without PVP tablets

Plasma profiles of rabbits after administration of ceftriaxone (CTX+CDC+MCC+PVP) tablet

Mean plasma profiles of ceftriaxone formulations in rabbits

Table 1 : Granulation tests and preliminary stability assessment (6 months at 40°C/75%RH) of granules compressed into tablets

Granulation technique	Composition of granules	Flowability (s)	Bulk /Tapped D250 density (mg/ml)	CTX Assay after 6 months at 40°C/75%RH (% of T0)
fluidized bed granulation	CTX pure	21	0.47/0.53	NA (99.2% after 1 month)
	CTX+PVP 1%	40	0.42/0.63	101.7
	CTX+NaCDC	26	0.43/0.49	99.4
	CTX+NaCDC +2%PVP	17	0.42/0.55	102.6
traditional granulation	CTX+NaCDC +2%PVP	18	0.43/0.45	93.7
	CTX+NaCDC +5%PVP	32	0.54/0.59	91.0
	CTX+NaCDC +7%PVP	16	0.49/0.54	96.0
	CTX+NaCDC +10%PVP	17	0.49/0.51	92.9
dry granulation	CTX	16	NA	100.8
	CTX+NaCDC	16	0.62/0.64	90.0

NA : not available

Table 2 : Optimization of intra-granular and extra-granular phases of recto-dispersible tablet formulation

Intra-granular phase / quantity (mg)															
Ceftriaxone	API	500							500						
NaCDC	Absorption Enhancer	125							125						
Dicalcium phosphate (Emcompress)	Fillers	220													
MCC (Avicel pH 102)									220						
Mannitol										220			220	220	220
Corn starch											220				
Lactose monohydrate												220			
Pregelatinized corn starch (Sepistab)	Binders												10		
HPMC 50														10	
PVP K30		10							10	10	10	10			
Maltodextrin															10
Extra-granular phase / quantity (mg)															
HPMC 50	Bioadhesive	120	50	50	25	20	20	20	20						
Croscarmellose	Super-Disintegrants	30	30	50	50	50	70								
Crospovidone								70	70						
Aerosil /Mg Stearate (1/1/1)	Glidants / Lubricants	18							18						
Total (mg)		1023	953	973	948	943	963	963	963	963	963	963	963	963	963
Flowability of the grain (s)		NA	NA	NA	NA	NA	NA	16.5	26	16	infinite	24	20	18	17
Disintegration of tablet (min)		10	8	7	5	5	3	2	3	1.7	5	3	0.6	1.1	1.2
TD ₈₀ of tablet (min)		NA	NA	NA	NA	NA	NA	9.3	5	4	6.3	3.9	3.5	1.8	2.1

Bad preliminary stability indications*		NA	NA	NA	NA	NA	NA					X	X		X
Preliminary conclusion		X	X	X	X	X	X	Go	Go	Go	X	X Compression problem	X	High friability (>3%)	X

*browning of grain, HPLC demonstrated presence of degradation products.

Table 3: Dry granulation formulations

Composition (mg)	Dry granulation	
	TABLETS	
CTX	500	500
NaCDC	125	125
Mannitol	-	220
Dicalcium phosphate	220	-
PVP K30	10	10
HPMC 50	20	20
Crospovidone	70	70
Aerosil / Mg Stearate / Talc (1/1/1)	18	18
Evaluation		
Mass uniformity variation (%)	<5	<5
Friability (%)	4.4	>50
Hardness (N)	37.7	15.8
Disintegration (min)	6.4	3.1
Stability 40°C/75%RH after 6months / T0 (mean $\pm$ SD, %)	92.7	90.6

Table 4 : Final CTX forms evaluation with powder or wet granulated CTX and NaCDC

Composition (mg)	CAPSULES		TABLETS			
Formulation name	Powder filled capsule	Granulate filled capsule	Mannitol optimized tablet	MCC optimized tablet	MCC+PVP tablet	Calcium phosphate optimized tablet
CTX	500	500	500	500	500	500
NaCDC	125	125	125	125	125	125
MCC				230	220	
Mannitol			230			
Dicalcium phosphate						230
PVP K30					10	
HPMC 50			20	20	20	20
Crospovidone			70	70	70	70
Aerosil / Mg Stearate / Talc (1/1/1)			18	18	18	18
Evaluation						
Mass uniformity (%)	<5	<5	<5	<5	<5	<5
Friability (%)	-	-	2.12	1.15	3.20	1.03
Hardness (N)	-	-	43	43	28	55
Disintegration (min)	NA	NA	1.3	1.0	2.5	2.5
Stability 40°C/75%RH after 6months / T0 (mean ± SD, %)	94.4 ± 4.18	97.7 ± 1.47	91.1 ± 1.70	93.9 ± 0.51	92.5 ± 1.50	77.2 ± 14.40

NA : not available

Table 5 : Pharmacokinetic parameters of CTX rectal and IV formulations in rabbits

Ceftriaxone form (20mg/kg)	C_{max} (µg/ml)	T_{max} (min)	T (>MIC) (h)	AUC_{∞,ext} (µg*h/ml)	F (%)	F' (%)
IV solution (n=6)	203 ±70	-	11.2 ±4.2	281 ±92	100 ±33	-
Suppository (n=5)	28 ±23	6 ±2	4.4 ±1.4	56 ±24	20 ±8	100 ±43
Capsule containing powder blend (n=5)	26 ±18	18 ±7 (*)	3.6 ±2.5	47 ±33	17 ±12	85 ±59
Capsule containing granulates (n=6)	16 ±16	15 ±0 (*)	2.0 ±2.4	27 ±31	10 ±11	49 ±55
Tablet containing MCC and PVPK30 (n=4)	16 ±8	14 ±11	2.7 ±1.4	49 ±32	17 ±11	88 ±57
Tablet containing MCC (but no binder) (n=3)	15 ±7	8 ±6	2.2 ±2.0	30 ±20	11 ±7	54 ±35
Tablet containing mannitol (n=6)	18 ±8	7 ±4	3.3 ±2.3	42 ±26	15 ±9	76 ±46

(*) : significant difference of parameter compared to suppository (control) formulation (Student test, p<0.05)