Electronic supplementary information for: New biocompatible hydroxy double salts and their drug delivery properties

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1. Experimental details

1.1 HDS synthesis

All materials were procured from Sigma-Aldrich (Gillingham, UK) and used without further purification. $[Mg_{2.1}Zn_{2.9}(OH)_8]Cl_2{\cdot}yH_2O$ (MgZn-Cl) was synthesised by reaction of ZnO (0.5 g) with $MgCl_2{\cdot}6H_2O$ (2.5 g) in 5 mL deionised water. The mixture was stirred for 3 days at room temperature, and the product recovered by vacuum filtration. The resultant white powder was rinsed with deionised water, and then allowed to dry under vacuum at 40 °C.

$[Fe_{2.4}Zn_{2.6}(OH)_8]Cl_2{\cdot}yH_2O$ (FeZn-Cl) was prepared by reacting ZnO (0.5 g) with $FeCl_2$ (1.18 g) in the presence of KI (0.5 g) in 15 mL deionised water. Air was excluded from the vial. The mixture was stirred for 3 days at room temperature, and the product recovered by vacuum filtration. The FeZn-Cl system tended to oxidise easily when in contact with air, and to minimise this water was rapidly drained from the preparation using a large filter funnel before the solid was washed with a small amount of deionised water. The product was then dried in a vacuum oven in the presence of silica gel at 40 °C and 0 % humidity.

1.2 Intercalation reactions

Intercalation of the drug anions was achieved by combining 0.4 mmol of each HDS with 1.6 mmol of sodium naproxen. 10 mL of deionised water was added to the solid materials, and the mixture stirred at 60 °C for 3 days in a sealed glass vial. KI (0.2 mmol) was added to the mixture of FeZn-Cl and the guest anions to reduce HDS oxidation, and the reaction vessel was additionally flushed with nitrogen. The solid products were filtered under vacuum, washed with deionised water, and dried. The intercalates of the FeZn-Cl HDS were treated with extra care as described above.

Using a 1:4 HDS : naproxen molar ratio gave two replacement naproxen ions for every chloride ion in the HDS, and ensured that complete intercalation was achieved. Using a 1:3 molar ratio was also successful, but an HDS : naproxen ratio of 1:1 unsurprisingly led to incomplete replacement of the initial guest. We opted to work with an HDS : naproxen ratio of 1:4 in all subsequent experiments to ensure complete reaction. After reactions were finished, the left-over guest solution could be recovered, augmented with additional guest, and then used for further reactions, thereby avoiding wastage.

1.3 Scale up

The reactions in Sections 1.1 and 1.2 were scaled up to produce products on the 100g scale, in order to have sufficient material for tablet making. Scale-up syntheses were performed in sealed Schlenk flasks fitted with paddle stirrers. Reaction conditions and reagent amounts are detailed in Table S1.

Table S1. Details of the scale-up reaction conditions. All reactions were performed in water.

Material	Starting materials	Volume of solvent (mL)	Temperature (°C)	Time (days)
FeZn-Cl	ZnO (100 g) $FeCl_2$ (240 g) KI (166 g)	2500	RT	3
MgZn-Cl	ZnO (200 g) $MgCl_2 \cdot 6H_2O$ (700 g)	1000	RT	3
FeZn-Nap	FeZn-Cl (0.5 mol) Sodium naproxen (2 mol) KI (0.2 mol)	500	60	3
MgZn-Nap	MgZn-Cl (0.5 mol) Sodium naproxen (2 mol)	500	60	3

1.4 Tablet production

Tablets were prepared using the direct compression method. Microcrystalline cellulose (Avicel® PH101) and spray dried mannitol (Pearlitol®200) were used as fillers and compression aids. Polyvinyl pyrrolidone (PVP K44) and magnesium stearate were used as a binder and lubricant, respectively. Mixtures of powders were prepared as detailed in Table S2, passed through a 60 mesh sieve, and mixed thoroughly. The powder mixture was then compressed using a single punch tableting machine (F3, Manesty Machines Ltd, Liverpool, UK) fitted with a 10 mm flat-faced punch. This results in tablets containing a dose of approximately 50 mg of Nap with each HDS.

Table S2. The powder mixtures used to prepare tablets.

Sample	HDS-Guest (%)	(Avicel® PH 101) (%)	PVP K44 (%)	(Pearlitol® 200) (%)	Mg stearate (%)
MgZn-Nap-Tab	60.0	24.1	1.3	14.1	0.5
FeZn-Nap-Tab	58.1	26.9	1.0	13.2	1.0

2. Characterisation

2.1 Materials characterisation

The morphology of the samples was investigated using a scanning electron microscope (SEM; SUPRA 55, Zeiss, Oberkochen, Germany) with an accelerating voltage of 20 kV. The instrument was fitted with an energy dispersive X-ray (EDX) spectroscopy attachment for elemental analysis. C, H, and N contents were determined using the quantitative combustion technique on a CE1108 elemental analyser (Carlo Erba, Wigan, UK).

The particle size and zeta potential of MgZn-Cl and FeZn-Cl were recorded on a Zetasizer (Nano ZS, Malvern Instruments Ltd, Malvern, UK). Powder X-ray diffraction (XRD) patterns were obtained using a MiniFlex 600 diffractometer (Rigaku, Tokyo, Japan), using Cu Kα radiation at 40 kV and 15 mA. IR spectra were collected on a Spectrum 100 instrument (PerkinElmer, Waltham, MA, USA). Data were recorded from 4000 to 650 cm^{-1} at a resolution of 2 cm^{-1}.

Thermogravimetric analysis was carried out on a Discovery TGA instrument (TA Instruments, New Castle, DE, USA). The sample (*ca.* 5-10 mg) was mounted in an aluminium pan and heated at a rate of 10 °C min^{-1} between 30 °C and 400 °C under a flow of nitrogen (10 mL min^{-1}).

X-ray photoelectron spectroscopy was performed at the NEXUS facility (Newcastle University). A K-alpha instrument (Thermo Scientific, East Grinsted, UK) equipped with a monochromated Al Kα X-ray source was used with a pass energy of 40 eV and step size of 0.1 eV. Spectra were processed using the CasaXPS software (Casa Software Ltd., Teignmouth, UK).

2.2 Guest recovery

To ensure that naproxen could be recovered intact from its intercalation compounds, the latter (50 mg) were reacted with Na_2CO_3 (100 mg) in 5 mL of D_2O. The samples were filtered and NMR spectra collected on the filtrates using an AV-400 NMR spectrometer (Bruker, Billerica, MA, USA) operating at a ^1H frequency of 400.13 MHz. Samples of the pure drug were also dissolved in D_2O and analysed by NMR for comparison purposes.

2.3 Tablet specifications testing

2.3.1 Friability

20 tablets were pre-weighed and placed in a friability tester (TBH 200, Copley Scientific, Nottingham, UK). They were rotated 100 times, after which the tablets were recovered, any dust attached to them removed, and they were reweighed.

2.3.2 Hardness

The hardness of five tablets of each formulation was determined using a hardness tester (FR1000, Copley Scientific, Nottingham, UK).

2.3.3 Weight variation

20 tablets were weighed individually and the mean mass and standard deviation calculated.

2.3.4 Drug content

Four tablets of each formulation were finely ground using a mortar and pestle. The powder was transferred into a volumetric flask containing an acidic solution (HCl) and shaken for 2 h at room temperature. The suspension was then neutralised to pH 7 with NaOH and the final volume adjusted to 250 mL with deionised water. The solution was filtered, suitable dilutions were made, and UV absorbance values were recorded using a UV spectrophotometer (UV 1800, Shimadzu, Kyoto, Japan).

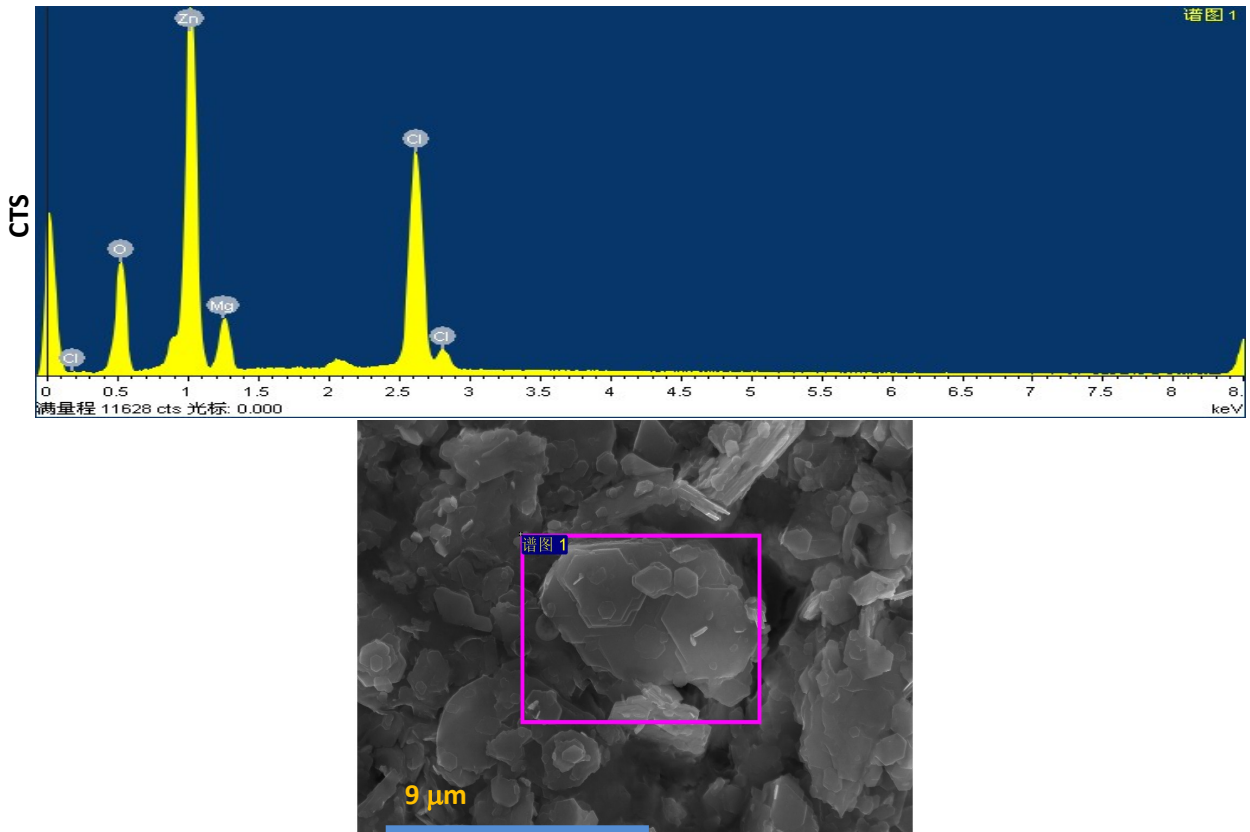
2.4 Functional performance tests

Drug release (dissolution) tests were carried out under experimental conditions as close as possible to the gastrointestinal tract and following pharmacopeia requirements.[1] The USP-II test (a paddle method) was used, with a PTWS instrument (PharmaTest, Hainburg, Germany) fitted with an inline spectrometer (CE 2500, Cecil, Cambridge, UK) being employed to perform these experiments. Drug release studies on the raw HDS-drug composites were carried out in 1 L of PBS (phosphate buffered saline) at pH 7.4, stirred at 50 rpm and held at 37 ± 0.5 °C. Tablets were initially placed in 750 mL of 0.1 M HCl in a vessel held at 37 ± 0.5 °C, and stirred at 50 rpm. After 2 hours of operation, the pH of the medium was adjusted to 6.8 ± 0.05 by adding 250 mL of 0.20 M tribasic sodium phosphate. Experiments were carried out for 22 h at this pH. Dissolution tests were undertaken in darkness, in triplicate with the new formulations prepared and 5 times with the commercial tablets.

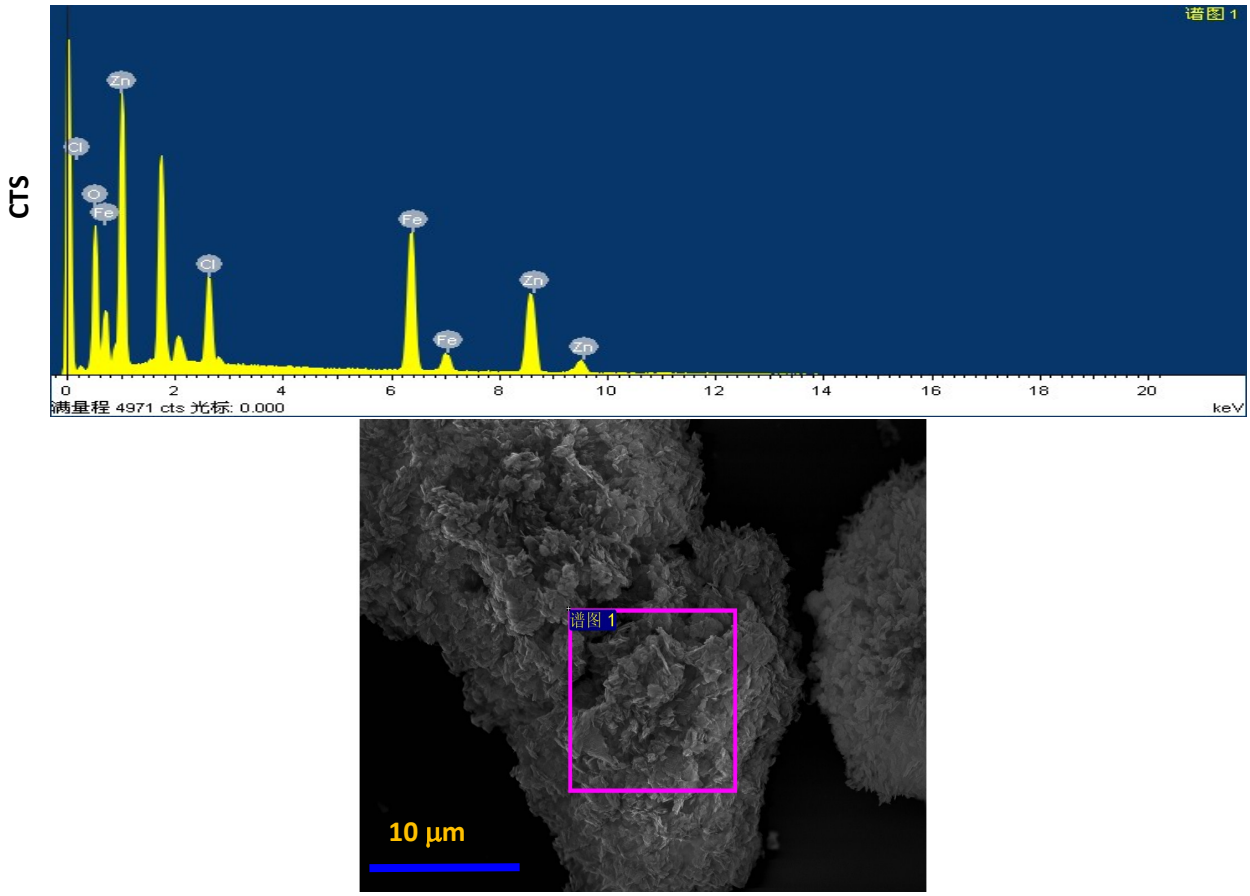
3. Results

3.1 Figures

Fig. S1. A digital photograph of the FeZn-Cl HDS.



(a)



(b)

Fig. S2. EDX data obtained on **(a)** MgZn-Cl and **(b)** FeZn-Cl. K and I show EDX signals at 3.312 and 3.937 keV; no such peaks can be observed in (b), demonstrating that there is no KI impurity present with the FeZn-Cl material.

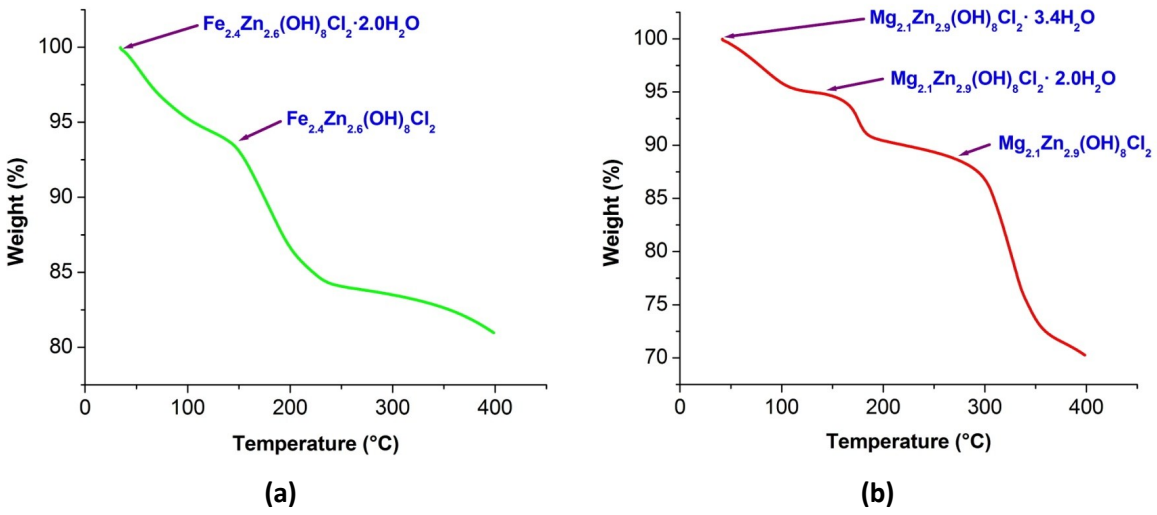


Fig. S3. TGA traces for **(a)** FeZn-Cl, and **(b)** MgZn-Cl.

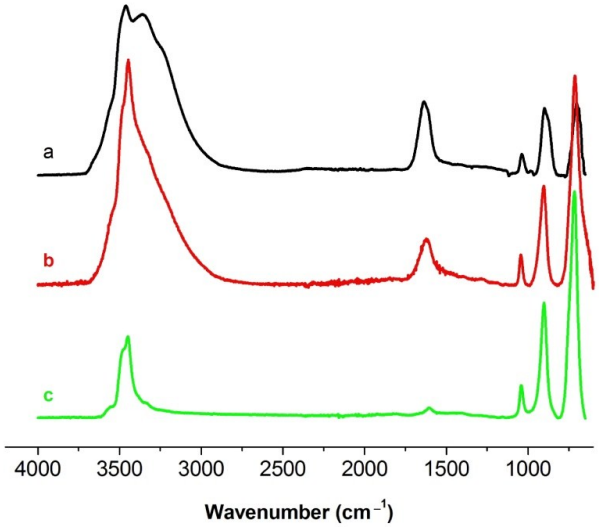


Fig. S4. IR spectra of **(a)** MgZn-Cl, **(b)** FeZn-Cl, and **(c)** Zn-Cl.

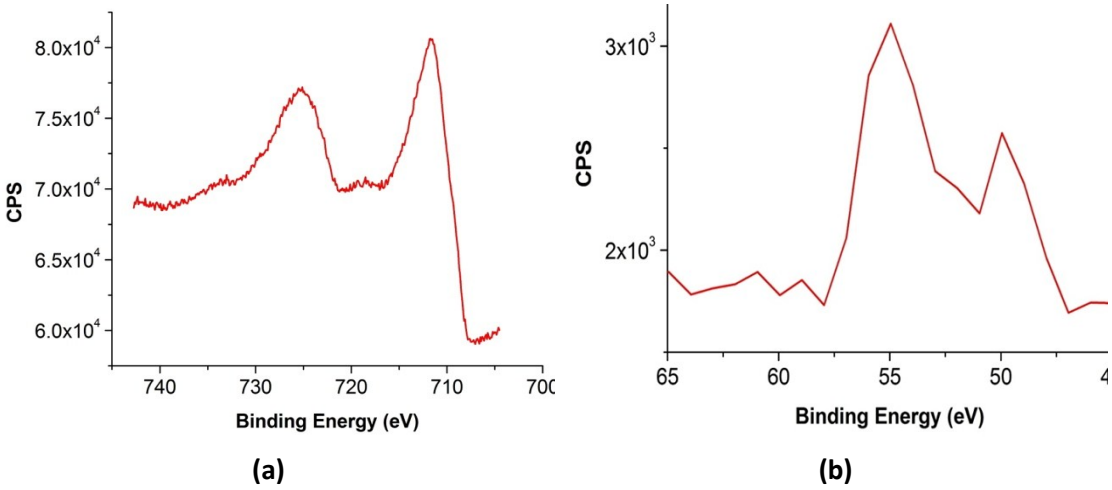


Fig. S5. XPS data for FeZn-Cl and MgZn-Cl depicting **(a)** the Fe 2p and **(b)** the Mg 2p regions.

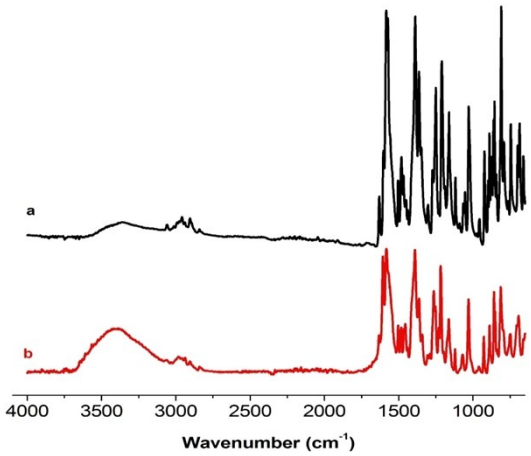


Fig. S6. IR spectra of **(a)** Nap and **(b)** MgZn-Nap.

3.2 Tables

Table S3. A summary of the particle size and zeta potential in water of the new HDSs.

	Size (nm)	Zeta Potential (mV)
FeZn-Cl	404 ± 239	45.1 ± 14.0
MgZn-Cl	360 ± 139	43.0 ± 13.9

Table S4. Indexing of the XRD pattern of the FeZn-Cl HDS. The initial cell used for refinement was that of $Zn_5(OH)_8Cl_2{\cdot}H_2O$.[2] Refinements were undertaken in the space group R-3m. The refined cell parameters are: a = 6.3443(16) Å; c = 23.6088(4) Å.

h	k	l	2θ(Obs) (°)	2θ(Calc) (°)	Difference (Obs – Calc) (°)
0	0	3	11.2704	11.2346	0.0358
1	0	1	16.5754	16.5524	0.023
1	0	4	22.0932	22.0857	0.0075
0	0	6	22.5991	22.579	0.0201
0	1	5	24.8595	24.8433	0.0162
1	1	0	28.1241	28.1075	0.0166
1	1	3	30.3527	30.3558	-0.0031
1	0	7	31.0865	31.0896	-0.0031
0	2	1	32.8118	32.7938	0.018
2	0	2	33.4759	33.4635	0.0124
0	1	8	34.4667	34.4698	-0.0031
1	1	6	36.387	36.3495	0.0375
2	0	5	37.8516	37.8588	-0.0072
1	1	9	44.7971	44.7971	0
0	0	12	46.0664	46.0997	-0.0333
2	1	4	46.311	46.3102	0.0008
1	2	5	47.8196	47.81	0.0096
3	0	0	49.7184	49.7445	-0.0261

Table S5. Indexing of the XRD pattern of the MgZn-Cl HDS. The initial cell used for refinement was that of $Zn_5(OH)_8Cl_2{\cdot}H_2O$.[2] Refinements were undertaken in the space group R-3m. The refined cell parameters are: a = 6.3200(37) Å; c = 23.8801(7) Å.

h	k	l	2θ(Obs) (°)	2θ(Calc) (°)	Difference (Obs – Calc) (°)
0	0	3	11.0238	11.1065	-0.0827
1	0	1	16.576	16.6036	-0.0276
1	0	4	22.0506	22.0145	0.0361
0	0	6	22.336	22.3191	0.0169
0	1	5	24.6976	24.7203	-0.0227
1	1	0	28.1701	28.2178	-0.0477
1	1	3	30.3685	30.4097	-0.0412
1	0	7	30.84	30.8609	-0.0209
0	2	1	32.9276	32.9167	0.0109
2	0	2	33.5794	33.5692	0.0102
0	1	8	34.1993	34.1878	0.0115
0	2	4	36.0585	36.0751	-0.0166
1	1	6	36.2918	36.2698	0.022
2	0	5	37.8652	37.8596	0.0056
1	0	10	41.2346	41.2115	0.0231
1	1	9	44.5612	44.5536	0.0076
2	0	8	44.9158	44.8986	0.0172
0	2	10	50.6682	50.6956	-0.0274

Table S6. Binding energies (in eV) of Fe 2p peaks and their satellites (SS) (see also Fig S5(a)). The Fe $2p_{3/2}$ peak is located at 712.0 eV and is associated with a small satellite peak at 718.8 eV. The binding energy (B.E.) difference between the Fe $2p_{3/2}$ peak and its satellite is 6.8 eV. For Fe^{2+} and Fe^{3+} the difference should be 6 and 8.5 eV, respectively.[3] The literature reports a B.E. difference of 8.5 eV for a Ca/Fe LDH containing iron in the Fe^{3+} oxidation state.[4] Thus, it appears that Fe in the FeZn-Cl HDS exists in the Fe^{2+} oxidation state with possibly a small amount of oxidation having taken place. There are not any XPS data reported for Fe^{2+} occupying octahedral and tetrahedral sites simultaneously; the only available data are for Fe^{3+}, but by comparing the FeZn-Cl data to Fe^{3+} XPS data[5] it is thought that Fe^{2+} occupies both octahedral and tetrahedral positions. This hypothesis is also in agreement with UV spectroscopy analysis (data not shown).

Material	Fe $2p_{3/2}$	SS	Fe $2p_{1/2}$	SS	References
FeO	710.1	715.1	723.8	730.0	[6,7]
Fe_2O_3	710.9	719.3	724.4	733.0	This work
$FeCl_2$	710.8	716.2	724.5	730.3	This work
FeZn-Cl	712.0	718.8	725.4	734.0	This work
$[Ca_2Fe(OH)_6]Cl{\cdot}2H_2O$	710.4	718.8	724.8	332.5	[4]

Table S7. Mg binding energies (in eV) determined from XPS spectra. For MgZn-Cl there are two peaks with shoulders, present in the area ratio of 1:1.9. The first is located at around 49.9 eV, similar to the values recorded for octahedral Mg^{2+} in the MgO structure and Mg/Al LDHs. The second peak at 55.0 eV is linked to Mg^{2+} in a tetrahedral site, as seen for $MgAl_2O_4$. This is indicative of Mg being in both octahedral and tetrahedral sites in the HDS.

Material	Mg 2p (eV	References
MgO	49.6	[8]
$MgAl_2O_4$	56.0	[9]
Mg/Al LDHs	49.7/49.9	[10,11]
MgZn-Cl	49.9/55.0	This work

Table S8. A summary of the drug release parameters for MgZn-Nap and FeZn-Nap in PBS (pH 7.4) and a commercial tablet formulation (Naprelan®). Please note that Naprelan is not the same as the Naprosyn discussed in the text (we were unable to source Naprelan for this work).

Time (h)	Drug Release (%)		
	FeZn-Nap	**MgZn-Nap**	**Naprelan®** [12,13]
0.5	14.3	37.0	25-60
1	23.8	51.2	35-75
4	60.0	80.8	> 65

Table S9. A summary of the results from US Pharmacopoeia specifications tests on the HDSs tablets. Values are reported as mean ± S.D., with the number of replicates for each experiment detailed below.[14-17]

Formulation	Weight uniformity n=20	Hardness (N) n=5	Friability (weight loss) (%) n = 20	Drug content (theoretical loading %) n=4
Pharmacopeia requirements	80-250 mg: ≤ 7.5 % different from mean >250 mg: ≤ 5.0 % different from mean	-	< 1.0	85 – 115 % of mean
MgZn-Nap-Tab	200.3 ± 3.0 mg *All tablets within 92.5 and 107.5% of the mean.*	123.0 ± 9.9	0.3 ± 0.08	100.9 ± 2.8 % *All tablets within 85 – 115% of the mean value.*
FeZn-Nap-Tab	331.5 ± 5.0 mg *All tablets within 95 and 105% of the mean.*	119.2 ± 7.6	0.4 ± 0.07	105.0 ± 3.4 % *All tablets within 85 – 115% of the mean value.*

4. References

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