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New biocompatible hydroxy double salts and their drug delivery properties

A. Y. A. Kaassis,[a] M. Wei[b] and G. R. Williams[a]*

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Two biocompatible hydroxy double salts (HDSs) were synthesised for the first time and loaded with active pharmaceutical ingredients. Drug release was studied from these intercalates, and sustained release observed. The HDS-drug composites were further formulated into tablets which were found to comply with pharmacopeia requirements for delayed release dosage forms.

HDSs are functionally and structurally similar to the widely explored layered double hydroxide (LDH) materials. These contain positive mixed metal hydroxide layers with charge balancing anions located between the layers. LDHs typically contain a mixture of divalent and trivalent metal ions.[1–3] The generic formula of an HDS is $[(M^{2+}_{2-x}Me^{2+}_{x})(OH)_{4-y}]X^{n-}_{y/n}\cdot zH_2O$, in which M^{2+} and Me^{2+} correspond to divalent metals ions such as Cu^{2+}, Co^{2+}, Ni^{2+}, or Zn^{2+} and X^{n-} is an exchangeable interlayer anion.[4,5] An example of an HDS is the zinc basic salt (ZBS) $[Zn_5(OH)_8](NO_3)_2\cdot 2H_2O$.[6–8] This comprise layers of edge-sharing $M(OH)_6$ octahedra with one third of the octahedral sites vacant and additional Zn^{2+} cations situated above and below the layer in tetrahedral sites. HDSs are generally stable and inert, and have been employed as biomolecule reservoirs or advanced green materials.[9] They can also be used as antimicrobial agents,[10] antifungal agents,[10] in water treatment,[11] as anticorrosion agents[12] and in photocatalysis.[13]

LDHs have been widely explored as drug delivery systems,[14–16] but HDSs have received much less attention in this regard with only a handful of reports in the literature.[17–20] The results that have been obtained with HDSs indicate that in general they release intercalated active pharmaceutical ingredients (APIs) in a more sustained manner than LDHs do.[21,22] However, there remain concerns regarding the accumulation of inorganic materials in the body upon repeated applications.[23] Hence, HDSs might be problematic if used in significant quantities to deliver APIs, should they break down during gastrointestinal (GI) passage. This is because they commonly contain toxic ions such as Cu^{2+} or Ni^{2+} and it is well known that free metals can be absorbed during their passage through the GI tract.[24] Zinc is an essential trace element in humans, so the ZBS system is less toxic than other HDSs. The LD_{50} for Zn chloride is around 350 mg/kg in rats when ingested orally. Thus, the ZBS would be safe if used in small amounts to deliver APIs, but could become toxic when higher amounts are used.[25–27]. In order to ameliorate this issue, here we report two new highly biocompatible HDSs constructed using Zn, Mg and Fe (the oral LD_{50} for $MgCl_2$ and $FeCl_2$ in rats are 2800 and 895 mg/kg, respectively). We further describe the incorporation of several APIs into their interlayer spaces, and the formulation of the resultant HDS-drug composites into tablets.

New HDSs made from the biocompatible metal ions Fe^{2+}, Zn^{2+}, and Mg^{2+} were prepared (as detailed in the ESI) using $FeCl_2$, $MgCl_2$ and ZnO as starting materials. HDSs were generated with mixtures of Fe/Zn, Mg/Zn, and Zn alone. These are henceforth denoted MZn-Cl (M = Mg, Fe) or Zn-Cl. The new materials were first thoroughly characterised to ensure HDS preparation was successful. FeZn-Cl (Fig. S1, ESI) has a dark green colour. This implies that the Fe^{2+} has not been oxidised to Fe^{3+}, and remains in the divalent oxidation state. Both MgZn-Cl and FeZn-Cl consist largely of hexagonal platelets (Fig. 1), in good agreement with the literature on HDSs.[28,29] The particle sizes were around 400 nm and positive zeta potentials were observed in water, as expected for an HDS structure (see Table S3, ESI).

X-ray diffraction (XRD) patterns (Fig. 2) show that MgZn-Cl and FeZn-Cl are indeed HDSs. Their patterns are very similar to that of Zn-Cl, showing both strong basal reflections (consistent with a layered structure) and a number of non-basal reflections (as a result of the ordered arrangement of ions in the layers). The MgZn-Cl and FeZn-Cl materials have slightly higher

[a] *UCL School of Pharmacy, University College London, 29-39 Brunswick Square, London WC1N 1AX, U.K.*
[b] *State Key Laboratory of Chemical Resource Engineering, Beijing University of Chemical Technology, Beijing 100029, P. R. China*
** Author for correspondence. Email: g.williams@ucl.ac.uk; tel: +44 (0) 207 753 5868.*

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interlayer spacings (8.1 and 7.8 Å respectively) than the Zn-Cl reference material, but can be indexed on very similar unit cells (full indexing is given in Tables S4 and S5, ESI).

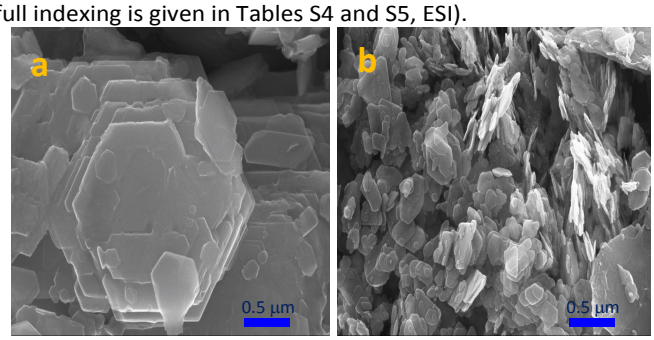

Fig. 1. Scanning electron microscopy images of (a) MgZn-Cl and (b) FeZn-Cl.

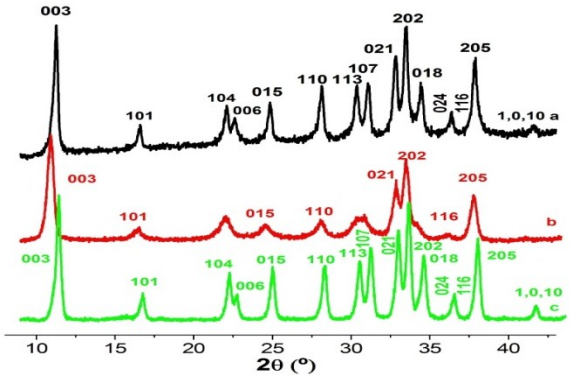

Fig. 2. XRD patterns of (a) FeZn-Cl; (b) MgZn-Cl; and, (c) Zn-Cl.

The chemical formulae of the new materials were calculated with the aid of energy dispersive X-ray (EDX) spectroscopy and thermogravimetric analysis (TGA) data. EDX analysis (Fig. S2, ESI) reveals the existence of a mixture of divalent metals (Fe/Zn and Mg/Zn) in the samples. From the metal ratios, the formulae of the new HDSs can be calculated as $Mg_{2.1}Zn_{2.9}(OH)_8Cl_2{\cdot}yH_2O$ and $Fe_{2.4}Zn_{2.6}(OH)_8Cl_2{\cdot}yH_2O$. In order to protect the Fe^{2+} from oxidation, KI was used during the synthesis of FeZn-Cl. The EDX data show that there is no KI present as an impurity in the FeZn-Cl product, since no peaks corresponding to either of these elements can be seen in the EDX spectrum.

Thermogravimetric analysis (TGA) traces are given in Fig. S3. Mass loss below 400 °C goes through two or three stages, which is common for LDH and HDS materials.[30–32] For FeZn-Cl, the initial mass loss of 6.64 % is complete by 160 °C and corresponds to the loss of two water molecules per formula unit (calcd. mass loss 6.59 %). With MgZn-Cl, the water mass loss of 12.24 % that commences below 100 °C and finishes at ca. 200 °C corresponds to a value of y = 3.4 (calcd. mass loss 12.04 %).

The IR spectra of MgZn-Cl, FeZn-Cl and Zn-Cl (Fig. S4, ESI) all look similar. MgZn-Cl and FeZn-Cl show broad peaks around 3450 cm^{-1}, which correspond to stretches of the OH groups in the layers and of interlayer water. In addition, a band around 1635 cm^{-1} is assigned to the δ-bend of water molecules in the interlayer. The M–O and O–M–O (M = Mg, Fe and Zn) vibrations appear at *ca.* 700 cm^{-1},[33] while peaks around 1030 and 900 cm^{-1} correspond to the bending of OH groups attached to divalent metal ions.

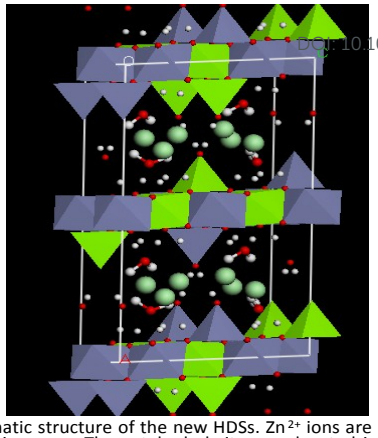

Fig. 3. A schematic structure of the new HDSs. Zn^{2+} ions are coloured purple and Mg^{2+}/Fe^{2+} in green. The octahedral sites are located in the layers and tetrahedral sites are situated above and below. Cl^- anions shown as cyan spheres, with water molecules (O red, H white) also visible in the interlayer space.

X-ray photoelectron (XPS) spectra were obtained on the FeZn-Cl HDS (Fig. S5) to investigate the oxidation state of Fe in the system. The data show that Fe overwhelmingly exists in the Fe^{2+} oxidation state (Table S6), with possibly a small amount of oxidation having taken place. The Fe^{2+} ions are believed to occupy both octahedral and tetrahedral positions. The Mg 2p XPS spectrum (Fig. S5 and Table tabS7) also demonstrates that Mg atoms are also likely occupy both octahedral and tetrahedral sites in the MgZn-Cl HDS. A schematic of the HDS structure is depicted in Fig. 3.

The active pharmaceutical ingredient naproxen sodium (Nap) was intercalated into both MgZn-Cl and FeZn-Cl using an anion exchange method (see ESI for details). Successful intercalation was confirmed by XRD, IR spectroscopy, and elemental microanalysis.

XRD patterns of the drug intercalates of MgZn-Cl are given in Fig. 4. The reaction products show no basal reflections characteristic of the starting material, and a shift of the 00l basal reflections to lower angles; this corresponds to an increase in interlayer distance, which is indicative of the incorporation of a larger anion into the interlayer galleries (see Table 1). The interlayer spacing increases from 8.1 Å with MgZn-Cl to 24.4 Å for MgZn-Nap; a similar increase from 7.8 to 24.5 Å is seen for FeZn-Nap. These expanded interlayer spacing values are in good agreement with the literature on Nap intercalates of LDHs and HDSs where d-spacings around 23.5 and 24.1 Å have been reported, respectively.[20,34,35] The diffraction patterns illustrate peak broadening, indicative of stacking defects. The elemental microanalysis results (Table 1) confirm the presence of drug, together with some residual Cl ions in the case of MgZn-Nap and some carbonate impurity (from dissolved CO_2 in the water used for reaction) in the FeZn-Nap material.

The IR spectra of the drug intercalates show all the characteristic peaks of Nap after intercalation (see Fig. S6, ESI, for the data on MgZn-Nap), confirming successful intercalation of intact API ions. Some of the peaks have shifted to slightly different wavenumbers due to interactions between the guest and host layers. The chemical integrity of the guest ions was verified through deintercalation and analysis by NMR (data not shown).

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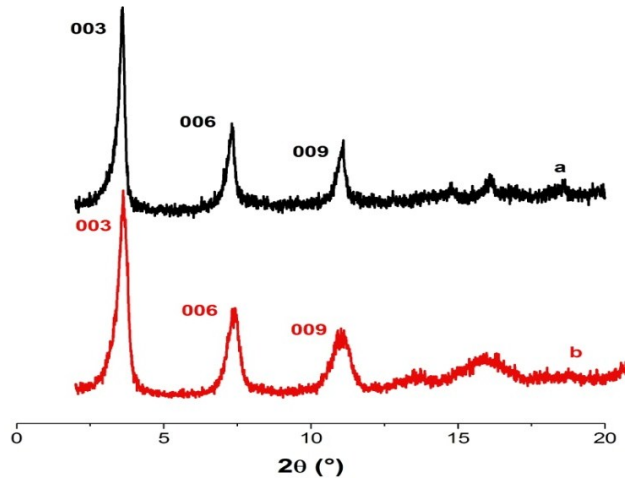

Fig. 4. XRD patterns for (a) FeZn-Nap, and (b) MgZn-Nap.

Table 1. The interlayer spacings and chemical formulae of the various MgZn-drug composites prepared.

ID	d_{003} (Å)	Formula	Elemental analysis (%) Obsd (calcd)[a]
MgZn-Nap	24.4	$Mg_{2.1}Zn_{2.9}(OH)_8(C_{14}H_{13}O_3)_{1.7}Cl_{0.3}\cdot 6.2H_2O$	C 35.41 (35.43) H 4.09 (4.45) H_2O 6.17 (6.20)
FeZn-Nap	24.5	$Fe_{2.4}Zn_{2.6}(OH)_8(C_{14}H_{13}O_3)_{0.8}(CO_3)_{0.6}\cdot 3.6H_2O$	C 19.57 (19.59) H 2.47 (3.39) H_2O 3.58 (3.60)

[a] C and H contents were determined by quantitative combustion, and the H_2O content from TGA

In vitro drug release studies of the Nap intercalates were undertaken in phosphate buffered saline (PBS; pH 7.4) at 37 °C; the results are depicted in Fig. 5. FeZn-Nap displayed slower release than MgZn-Nap. Release occurs over an extended period of time of ca. 1280 min (21.3 h); the HDS intercalates thus meet the pharmacopeia requirement for extended release, which requires the API to be freed over a prolonged period of time.[36] The difference in the release rate is thought to be related to the HDSs' metal compositions, since it is known that metals influence the release property from LDHs.[37,38] Release from the HDS systems reported here is much slower from most Nap-containing LDH systems (e.g. some Mg/Al LDH-Nap systems where 90 % of the drug was released after 4 h).[39,40] The range of values reported for LDH-Nap intercalates in the literature varies widely, however: Carriazo *et al.* and Gu *et al.* both explored Mg/Al LDH-Nap intercalates and found that 90 % of the incorporated drug was released after 35 min and 8 h respectively.[41,42]

The HDS-Nap composites were compared with literature data on drug release from a commercially available Nap tablet, and found to perform comparably with this (Table S8). To further investigate the drug delivery properties of the Nap intercalates they were formulated into tablets (MgZn-Nap-Tab and FeZn-Nap-Tab) using commercial excipients (see ESI for details). This required a scale-up of the initial reaction by a factor of 200-fold (details are given in the ESI). The materials resulting from scale-up were found to be identical to those prepared on the small-scale. The HDS tablets were tested for hardness, weight and content uniformity, and friability (see ESI Table S9) and in all cases found to meet the appropriate US Pharmacopoeia requirements.[43–45] Drug release tests comparing tablets of FeZn-Nap and MgZn-Nap with commercially available Naprosyn EC® (enteric-coated) tablets were undertaken, and the results are given in Fig. 6.

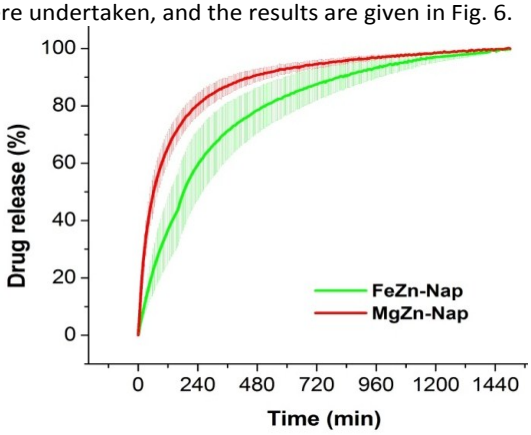

Fig. 5. Drug release from the Nap-loaded HDS powders at pH 7.4. Data from three independent experiments are shown as mean ± S.D.

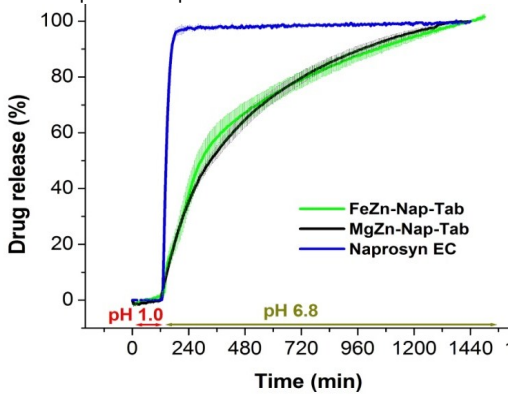

Fig. 6. Nap release from tablets of FeZn-Nap and MgZn-Nap at pH 1 and 6.8. Data from three independent experiments (5 for Naprosyn-EC) are shown as mean ± S.D.

These drug release tests were performed using pharmacopoeia standard protocols aiming to simulate the passage of the formulation through the human body after oral administration. The three formulations showed virtually no release during the first 2 h at pH 1 (mimicking the stomach). The HDS tablets are able to protect the drug from release as effectively as the commercial enteric coated tablet. Once the pH was adjusted to 6.8, release from the HDS tablets proceeded in a sustained manner (the EC tablet releases very rapidly at this pH, in order to deliver rapid relief of symptoms). Both HDSs display almost identical release profiles, and importantly show sustained release over *ca.* 24 h at neutral pH. The HDS tablets meet the US Pharmacopoeia requirements for delayed/extended release dosage forms: these specify that less than 10 % of the incorporated drug should be released in the acidic media and the drug is freed over a prolonged duration of time, respectively.[36,46]

In conclusion, two new biocompatible HDSs based on Mg/Zn and Fe/Zn were successfully synthesised for the first time. They comprise hexagonal platelets, and XPS data suggest that Mg^{2+}

and Fe^{2+} occupy both octahedral and tetrahedral sites. Bioactive guests including naproxen (Nap) have been successfully intercalated into the new HDSs. NMR and IR spectroscopy showed that the guest ions' structures remained intact after intercalation. Nap release in phosphate buffered saline (pH 7.4) was explored, and the drug-loaded HDSs showed extended release profiles. As a result of these promising results, HDS synthesis was scaled up to the 100s of grams scale and tablets were prepared with the HDS-Nap systems. The tablets produced meet all appropriate US Pharmacopoeia requirements both for the physical properties of the tablets and as delayed or extended release systems. HDS-based tablet formulations hence offer a novel platform for the design of future dosage forms.

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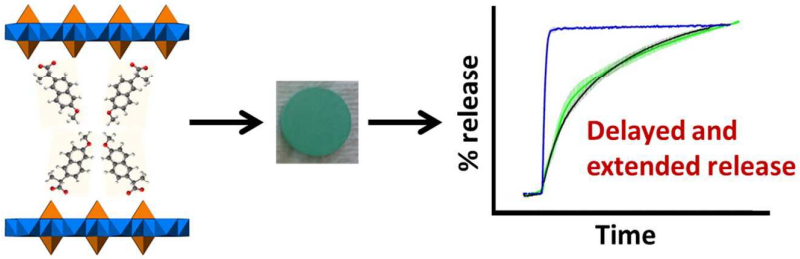
Graphical abstract for:
New biocompatible hydroxy double salts and their drug delivery properties

A. Y. A. Kaassis,[a] M. Wei[b] and G. R. Williams[a]*

[a] UCL School of Pharmacy, University College London, 29-39 Brunswick Square, London, WC1N 1AX, UK.

[b] State Key Laboratory of Chemical Resource Engineering, Beijing University of Chemical Technology, Beijing 100029, P. R. China

* Author for correspondence: E-mail: g.williams@ucl.ac.uk; Tel: +44 (0)207 753 5868.



Two novel biocompatible hydroxy double salts (HDSs) have been synthesised, loaded with the drug naproxen, and formulated into tablets.