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Review

The Advantages of Polymeric Hydrogels in Calcineurin Inhibitor Delivery

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Abstract: In recent years, polymeric hydrogels (PolyHy) have been extensively explored for their applications in biomedicine as biosensors, in tissue engineering, diagnostic processes, and drug release. The physical and chemical properties of PolyHy indicate their potential use in regulating drug delivery. Calcineurin inhibitors, particularly cyclosporine (CsA) and tacrolimus (TAC), are two important immunosuppressor drugs prescribed upon solid organ transplants. Although these drugs have been used since the 1970s to significantly increase the survival of transplanted organs, there are concerns regarding their undesirable side effects, primarily due to their highly variable concentrations. In fact, calcineurin inhibitors lead to acute and chronic toxicities that primarily cause adverse effects such as hypertension and nephrotoxicity. It is suggested from the evidence that the encapsulation of calcineurin inhibitors into PolyHy based on polysaccharides, specifically alginate (Alg), offers effective drug delivery with a stable immunosuppressive response at the in vitro and in vivo levels. This not only may reduce the adverse effects but also would improve the adherence of the patients by the effective preservation of drug concentrations in the therapeutic ranges.

Keywords: polymeric hydrogels; drug delivery; calcineurin inhibitors; cyclosporine; tacrolimus

1. Introduction

Hydrogels represent a special class of polymers that are widely found in nature, fulfilling important functions in tissues such as elastin or collagen. Hydrogels are three-dimensional hydrophilic polymers forming cross-linked networks. They possess fascinating physical and chemical properties and adopt different shapes and sizes depending on the existing environment.

Recently, PolyHy have aroused great interest in different disciplines of science due to their biocompatibility and ability to absorb water. Relative to other polymeric materials, these properties make them excellent candidates for applications in biomedicine, such as the construction of diagnostic devices, cell culture substrates, blood detoxifiers, tissue engineering, drug delivery systems, etc. This review gives an overview of the classification and characterization of PolyHy, focusing on their physical and chemical properties for their application in controlled drug release.

1.1. Family of Hydrogels

Polymeric hydrogels can be classified based on different factors:

i. Types of bonds in the reticulated three-dimensional structure:

- Physically cross-linked PolyHy: This hydrogel undergoes a transition from the three-dimensional state to the initial state of polymeric chains in solution and thus constitutes the so-called 'reversible ones'. Cross-links are formed via attractive forces or interactions between the polymeric chains (Figure 1A), such as hydrogen bonding, hydrophobic and electrostatic interactions [1]. This group consists of multifunctional hydrogels based on the copolymers of polyacrylamide and tannic acid that are cross-linked through hydrogen bonds [2].
- Chemically cross-linked PolyHy: This type of hydrogel is more stable in comparison with the physically cross-linked one due to the involvement of covalent bonds (Figure 1B). This indicates that they are irreversible, unless labile covalent bonds are intentionally introduced into the cross-linked structure. For instance, the chemical cross-linking of poly(vinyl alcohol) (PVA) hydrogels with citric acid has been successfully obtained to generate a controlled drug release system. Citric acid is a cheap and non-cytotoxic reagent compared to other crosslinkers that cannot be employed in biomedical applications due to their toxicity [3].

ii. Stimuli-responsive hydrogels. The advances in chemical synthesis, availability of new precursors, and improvement in reaction mechanisms have resulted in the development of biomaterials that respond to environmental stimuli. This promotes the conversion of static hydrogels to dynamic ones that respond to generated changes or specific inputs in variables such as temperature and pH. Furthermore, the stimulus-sensitive PolyHy are known as 'smart hydrogels' because of their response to any slight changes in the environment, such as temperature, pH, ionic strength, light, electric field, and biomolecules, by changing the degree of swelling. The response of the smart hydrogels depends on the type of monomer or polymer used and/or structural modifications within the compound. The challenges in the development of smart hydrogels that exhibit a rapid response to the stimuli of the medium are primarily based on synthetic mechanisms [4–9].

iii. PolyHy can also be classified based on their charge. Hydrogels are composed of molecular structures such as nonionic, cationic, anionic, amphoteric polyelectrolytes, or zwitterionic species. The development of zwitterionic hydrogels further confirmed their potential application in wound dressing [10,11]. The dual cross-linked networks composed of anionic and cationic monomers improve the mechanical properties of hydrogels and thus find application as injectable hydrogels [12]. On the other hand, new synthetic methodologies have been introduced in the field of cell adhesion to obtain composite hydrogels incorporating cationic groups in their structure to improve flexibility as well as cell adhesion [13].

iv. Another classification involves natural and synthetic hydrogels. Due to the biocompatible, biodegradable, and non-toxic nature, natural hydrogels find potential biomedical applications. Based on the type of natural polymer, there are three biomaterials, namely protein-based, polysaccharide-based, and natural polyester-based hydrogels [14–17]. In this review, we are interested in focusing on the biomedical applications of hydrogels based on polysaccharides, mainly. Therefore, later, we dedicate a section to this type of biomaterial. On the other hand, synthetic hydrogels have favorable properties for industrial applications, such as superior mechanical properties compared to natural hydrogels. However, hybrid hydrogels with improved and intermediate properties between natural and synthetic hydrogels have been manufactured. Biocompatible hydrogels functionalized with carbon nanotubes have been successfully synthesized to stabilize hydroxyapatite. The cytotoxicity results of these materials show promising insights for their application in bone tissue engineering [18]. In addition, there is a group of biodegradable synthetic hydrogels, which have been widely used for biomedical applications. Principally, in this group we can mention polymers, such as poly(caprolactone), poly(glycolic acid), poly(lactic acid), poly(D,L-lactide-co-glycolide), and polyurethane [19]. The studies on fucoidan-modified PVA hydrogels have revealed substantial improvement in cell

adhesion and hemocompatibility, suggesting the utility of these hybrid materials as implants or vascular devices [20]. In cell microenvironment engineering, these materials provide very thin platforms that are utilized to seed the endothelial cells. An attractive biocompatible scaffold based on modified gelatin and a biodegradable polyester of poly(D,L-lactic acid) (PDLLA) was developed. The combination of these two biomaterials provides the ideal environment for cytocompatibility and cellular interaction exhibited by gelatin. In addition, PDLLA provides appropriate mechanical resistance for the graft tissue in corneal endothelial transplant [21].

v. Based on their size, hydrogels are classified into those containing macroscopic structures and those containing networks of smaller dimensions, such as microgels or nanogels. The conformation of hydrogels with micrometric precision at the level of individual cells has allowed their application as artificial cells [22,23], drug and cell carrier systems [24,25], and assembled elemental units in artificial tissue construction [26,27]. Polysaccharide-based microgels are one of the most commonly used hydrogels. Recent studies have focused on microgels that can stabilize and release lipophilic particles. Microgels were prepared from corn starch via an oxidized-annealing process. Further, the adsorption/release capacity of lysozyme by the microgels was evaluated. Studies showed that the oxidized-annealed microgels exhibit high charge and release capacities over a wide range of pH and ionic strength values. This is attributed to the electrostatic forces between the carboxyl groups generated by the oxidation-annealing process of the microgel and lysozyme [28]. The hydrogels embedded in microgels to form a hydrogel/microgel system have been utilized as wound dressings. The carboxymethyl chitosan- and oxidized carboxymethyl cellulose-based microgels are drug-loaded via the Schiff base reaction. The gel time, morphology, swelling ratio, weight loss ratio, mechanical properties, release profiles of pH-sensitive drugs, and antibacterial activities were analyzed. The addition of microgels was found to offer stability, improved mechanical performance, and sensitivity to drug release at different pH conditions for the hydrogels. The addition of Ag compounds to the hydrogel/microgel composites demonstrated desirable antibacterial properties, which ensured their possible application in wound dressing [9]. On the other hand, nanogels, which are also known as nano-sized hydrogels or hydrogel nanoparticles, are nanostructured three-dimensional networks composed of functional polymers, whose sizes are in the sub-micrometer region, typically from 20 to 200 nm. Hydrogel nanoparticles are formed by physical as well as chemical cross-linking methods. Nanogels offer several advantages, such as biocompatibility, high swelling capacity, and high-water solubility. In addition, they are classified as stimuli-responsive and nonresponsive hydrogels. Nanogels respond to fluctuations in environmental factors, such as temperature, pH, pressure, electric and magnetic fields, molecular species, ionic strength, or a combination of different factors. Therefore, nanogels are exceptional candidates for application in biomedicine, such as biosensors, drug release, tissue engineering, diagnosis, etc. [20,29,30].

Some of the examples of PolyHy are listed under the respective classifications in Table 1.

As mentioned in the previous classification summarized in Figure 2, PolyHy covers a wide range of materials and synthetic methods. In fact, these materials can be potentially applied in biomedicine due to their versatility to modify and adapt as required. This review focuses on naturally occurring, i.e., biodegradable, specifically polysaccharide-based hydrogels. Initially, the latest studies on natural hydrogels for application in tissue engineering are reviewed. The later sections present the studies on the controlled release of drugs by mainly focusing on calcineurin inhibitors (CNIs), which are common immunosuppressive drugs used in the treatment of autoimmune diseases and solid organ transplantation.

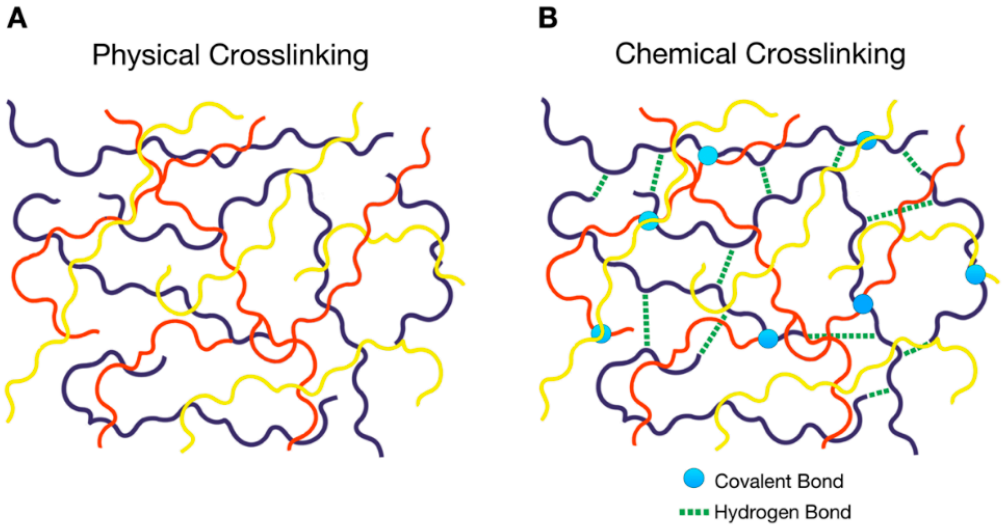

Figure 1. Schematic representation of the types of cross-linking in a polymer hydrogel (PolyHy); (**A**) physical cross-linking of different types of polymeric chains denoted by different colors, (**B**) chemical cross-linking consisting of covalent bonds between the polymeric chains denoted by circles and non-binding interactions such as hydrogen bonds represented by dashed lines.

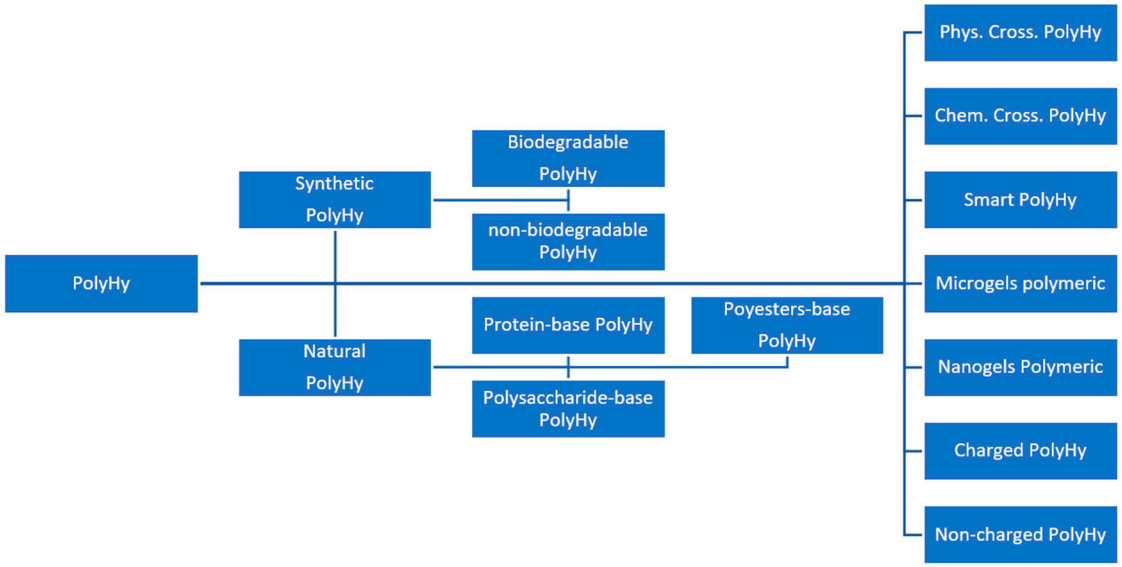

Figure 2. Polymeric hydrogels classification.

Table 1. Examples of polymeric hydrogels listed under respective classifications based on cross-linking, ionic charge, size, and origin.

Polymeric Hydrogels	Reference
Physically and chemically cross-linked	
Polyacrylamide/nanochitin	Dongjian Li. 2020 [31]
Poly methylcellulose	Niemczyk-Soczynska. 2019 [32]
Polycaprolactone	Cernadas. 2019 [33]
Gelatin methacryloyl/carrageenan	Lauren M. 2018 [34]
Poly(vinyl alcohol)-g-poly(*N*-isopropylacrylamide)	Sosnik A. 2017 [35]
Stimuli-responsive or smart hydrogels	
Polyacrylamide/polyethoxysiloxane	Dongjian Li. 2020 [31]
Polyhydroxyethylmethacrylate/N-isopropylacrylamide	Huang, H. 2020 [36]
Pluronic	Shriky, B. 2020 [37]
Chitosan/poly(allylamine hydrochloride)	Ghaffar, A. 2020 [38]
N-isopropylacrylamide/hydroxyethyl methacrylate/N,N-dimethylacrylamide	Wei, HL. 2020 [39]
Glucuronoxylan	Hussain, MA. 2020 [40]
Chondroitin sulfate-co-poly(acrylic acid)	Ahmad, M. 2019 [41]
According to their charge	
Poly(sulfobetaine acrylamide)/poly(lysine acrylamide)	Huang, CJ. 2020 [42]
Alginate/poly [2-(methacryloyloxy)ethyl]dimethyl-(3-sulfopropyl)ammonium hydroxide-co-acrylamide	Wang, ZK. 2020 [43]
Polydopamine	Huang, H. 2020 [36]
Poly(sulfobetaine methyl methacrylate)	Ramsey, JD. 2020 [44]
Sodium alginate/poly acrylic-acrylamide	Huang, H. 2020 [36]
Natural and synthetic hydrogels	
Natural soil/Alginate/polyacrylamide	Li, JC. 2020 [45]
Gelatin/gamma-polyglutamic acid	Liu, B. 2020 [46]
Gellan gum/lignocellulose	Kouhi, M. 2020 [16]
Chitosan/gamma-cyclodextrin/Clinoptilolite	Tonelli, AE. 2020 [47]
Arabinoxylan/guar gum	Haider, S. 2020 [48]
Polyglycolic acid/poly-lactic-co-glycolic acid	Bubbly, SG. 2020 [49]
Polyacrylamide/starch	Balaban, RD. 2020 [50]
Poloxamer 407	Priddy, LB. 2020 [51]
Microgels and nanogels polymeric	
Janus microgels	Ramakrishnan, S. 2020 [52]
Alginate microgels	Damiati, S. 2020 [53]
Poly(n-isopropylacrylamide) microgel	Holian, A. 2020 [54]
Poly(N-isopropylacrylamide)/polyethylene glycol/graphene oxides	Zhang, XQ. 2020 [55]
Chitosan/laponite nanogels	Kokabi, M. 2020 [56]
Poly(glycidol) nanogels	Albrecht, K. 2020 [57]

1.2. Tissue Engineering Applications of Natural Polymeric Hydrogels

The hydrogels consisting of natural polymers have been recognized as prime candidates for designing tissue scaffolds. This is attributed to the excellent characteristics such as cost efficiency, high compatibility, and biodegradability exhibited by this type of hydrogels. Natural polymeric hydrogels, such as polysaccharides and proteins, were similar to the natural extracellular matrix. Thus, the hydrogels are endowed with magnificent properties, such as cellular proliferation and survival, to overcome certain challenges in tissue engineering [58–60]. Recently, a microbial polysaccharide-based macroporous hydrogel was manufactured from salecan and gellan gum by a one-step method [58]. These were obtained via the association of polysaccharide chains by means of Van der Waals forces, hydrogen bonds, and hydrophobic interactions. It should be noted that the manufacturing methodologies did not require crosslinkers or toxic monomers. Instead, the salecan/gellan gum hydrogel that exhibits excellent biocompatibility and properties to mimic the native tissues were utilized. In this context, cardiac tissue engineering has emerged as a branch of tissue engineering, which uses combinations of cells, biological and/or synthetic materials, growth factors, differentiation factors, proangiogenic factors, and monitoring, as a promising and challenging approach to induce the

regeneration of heart tissue [61]. This field of research gave rise to the study of biocompatible hybrid systems [62], such as polysaccharide-based PolyHy, for application in tissue engineering [63,64].

Alginate (Alg) and chitosan (CS) are natural polysaccharides that are widely used in biomedicine, as well as food and pharmaceutical industries [65,66]. This is attributed to the high biocompatibility and biodegradability of polysaccharides [15,67]. A recent study on the improvement of the mechanical tensile strength of decellularized extracellular cardiac matrix (ECM) using Alg/CS hydrogels [68] showed a high swelling capacity, good mechanical resistance, moderate biodegradable properties, and excellent cell viability. The results of the study reveal that CS- and Alg-based platforms find potential applications in cardiac tissue engineering. In addition, other natural polymers, such as collagen, gelatin (Gel), and fibrin, have been studied for myocardial tissue engineering. The collagen/CS composite scaffold has been widely used for myocardial tissue engineering [63]. Since collagen, which is the main component of the ECM within the myocardium, has a fast degradation rate and poor mechanical properties [69,70], it has been combined with other polysaccharides to improve the characteristics of the composite PolyHy.

The research in tissue engineering aims to manufacture materials that mimic native tissues of the human organs. The biomimetic materials capable of mimicking the structure and composition of the native extracellular matrices are essential for successful organ tissue repair and transplantation in humans. Similarly, fibrin-agarose tissue-like hydrogels (FATLHs) have allowed successful biofabrication of different biological substitutes, and thus, delivered promising ex vivo and in vivo results [71]. These fibrin-agarose-based materials biodegrade and integrate into implanted areas and vital organs without any histopathological alteration. Thus, FATLHs exhibit potential clinical uses in engineering applications due to their biosecurity and biocompatibility. Furthermore, these artificial tissues combine to be stronger, and thus offer a stable mechanical response along with favorable cytocompatibility [72].

1.3. Advantages of PolyHy in Controlled Drug Distribution

It is quite interesting that the number of papers published based on the application of PolyHy in the controlled release of drugs in the journals indexed within the WoS database exceeded eight hundred articles in the last five years (Figure 3). Polymers have been widely used as biomaterials for the manufacture of medical devices as well as tissue engineering platforms. In particular, hydrogels have aroused great interest in biomedicine, mainly focusing on the development of scaffolds that release therapeutic agents [73–76]. In this context, hydrogels based on natural polymers have been extensively studied for applications in biomedicine due to their intrinsic characteristics of biocompatibility, biodegradability, and similarities to the extracellular matrix [45,77–79]. The physicochemical characteristics of biopolymeric hydrogels used in drug delivery systems, namely molecular weight, solubility, morphology, hydrophilicity, hydrophobicity, loading and release efficiency of the drug, surface energy, gelation process, etc., have been investigated [80–82].

Due to their natural abundance, biocompatibility, and physicochemical properties, hydrogels based on polysaccharides, specifically Alg and CS, are potentially used as drug delivery systems. Alg is a naturally abundant anionic polysaccharide extracted from brown algae, comprising two monomeric units, namely α-L-guluronic acid (G) and β-D-mannuronic acid (M) residues [83]. Due to the characteristic properties of Alg, such as affordability, non-toxicity, high adsorption capacity, and gelation capacity, these hydrogels are suitable for application in the food industry and biomedicine [84,85]. Alg hydrogels are prepared by physical or chemical cross-linking methods, as described above. However, the poor mechanical properties are a major drawback for the Alg hydrogels [86]. To resolve this limitation, previous studies reported the development of hybrid hydrogels. Jahanban-Esfahlan et al. [87] developed a hybrid hydrogel through the Schiff base con densation reaction of Alg with Gel, followed by the incorporation of Fe_3O_4 magnetic nanoparticles as a strategy to improve the mechanical properties of the hybrid hydrogel (Figure 4A). Scanning electron microscopy (SEM) was used to verify the microporous structure of Alg/Gel hydrogel without microphase separation (Figure 4B). The hydrogel containing Fe_3O_4 nanoparticles was loaded with an anti-cancer

drug, namely doxorubicin hydrochloride. The in vitro study revealed that the doxorubicin-loaded Alg-Gel/Fe_3O_4 magnetic hydrogel showed pH-responsive drug release behavior. Thus, the fabricated magnetic PolyHy has excellent potential as a drug delivery system for cancer chemotherapy.

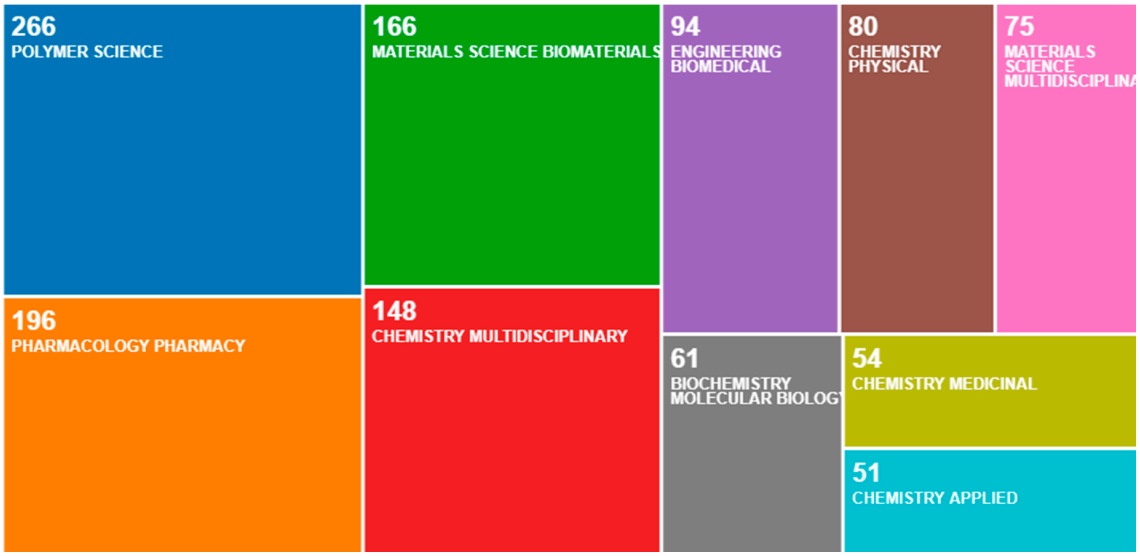

Figure 3. Tree map shows the number of articles published under different research areas in the last five years. The keywords used for the search and analysis were 'PolyHy' and 'Drug Releasing Systems'. WoS source, August 2020.

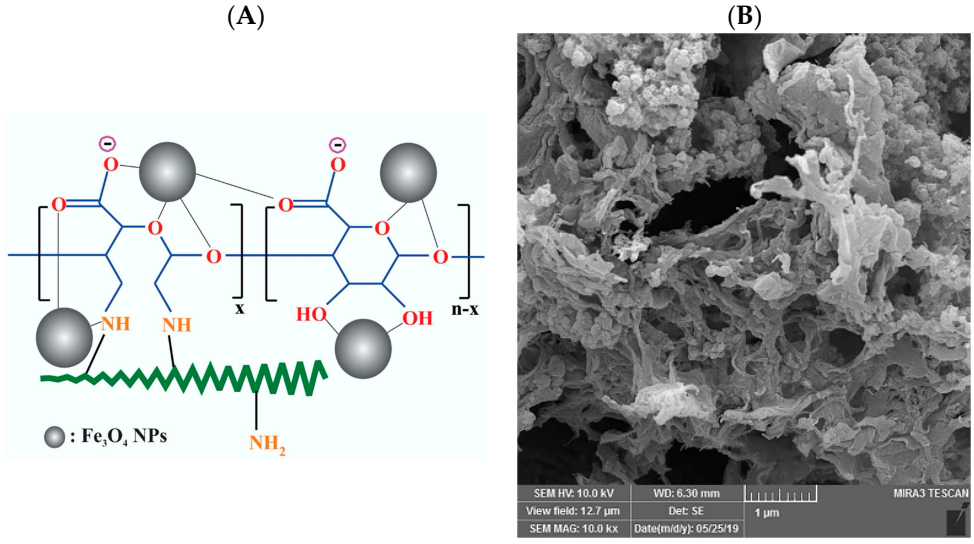

Figure 4. (**A**) The structure of a hybrid hydrogel containing Fe_3O_4 magnetic nanoparticles, and (**B**) microporous structure determined by SEM. The images were extracted and adapted from reference [87].

The recent studies on Alg-based hydrogels primarily focused on improving the mechanical properties [58,88], manufacturing methods [89,90], payload of gels [21], and transport and release of hydrophilic and hydrophobic drugs [91,92] such as CNIs. These Alg-based PolyHy are good drug delivery systems for the brain, cancer, antibacterial activity, immunosuppressive therapies, etc. [93–96]. This review is specifically focused on the studies undertaken for the transport and release of CNIs to find the optimal systems in order to minimize the side effects on the human body.

2. Calcineurin Inhibitors

2.1. Presentation, Classification, and Use

Calcineurin is a serine-threonine phosphatase with a calmodulin-binding catalytic subunit and a calcium-binding regulatory subunit. Due to its significance in cell development, this enzyme is expressed in most cells [97]; however, it has been better described in immune response. Particularly, the dephosphorylation of the transcription factor, namely the nuclear factor of activated T cells (NFAT), by calcineurin leads to translocation of the protein into the nucleus, which further activates different leukocyte proliferation and differentiation mediators [98]. This molecular mechanism is reported in T-lymphocytes, which are crucial for adaptive immune response and post-transplant organ rejection.

CNIs, as previously defined, are immunosuppressant drug-targeting calcineurins that are commonly used for the treatment of autoimmune diseases and subsequent to transplantation, including kidney transplantation. Furthermore, CNIs have been used for the treatment of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection [99]. CNIs include cyclosporine A (CsA) and tacrolimus (TAC), which binds cyclophilin and FK506 binding protein 12 (FKBP12), respectively [100]. These CNIs primarily inhibit the pathway of NFAT and thus prevent the transcription of interleukin-2 (IL-2), one of the most important cytokines for the growth and differentiation of T-helper lymphocytes.

Since their introduction in the 1970s, CNIs have significantly lower rejection rates, which further extends the time of graft survival. However, long-term side effects, such as nephrotoxicity [101] and hypertension [102], represent a current concern. On the other hand, CNIs were found to be safe in children diagnosed with glomerulopathy, which cannot be treated by steroids [103], another class of immunosuppressant drugs.

2.2. Pharmacological Aspects of CNIs

CNIs are important for immunosuppression in solid organ transplanted patients. They have a narrow therapeutic index that promotes therapeutic drug monitoring (TDM) in order to avoid progressive toxic effects over time, such as nephrotoxic, cardiovascular, or neurological side-effects. In addition, the introduction of suboptimal concentrations of CNIs reduces the risk of organ rejection [104]. TDM is performed routinely by the immunoassay of whole blood samples due to the distribution of around 90% TAC and 50% CsA on erythrocytes [105]. A trough blood sample is required for the accurate estimation of the area under the curve for TAC, while a sample extracted two hours after oral administration (C2) is used to obtain the TDM of CsA.

Due to high inter- and intra-variabilities in pharmacokinetics (PK), a sensitive equilibrium needs to be maintained after the administration of a dose of CNI. This variability is mainly explained by the pharmacogenetic influence of the cytochrome P450 system (hepatic and intestinal polymorphisms in CYP3A4 and CYP3A5, respectively) that alters the pharmacokinetic metabolism of these drugs [106,107]. In addition, the intestinal efflux-pump P-glycoprotein contributes to the PK variability, resulting in the reduction in drug absorption and change in disposition [108]. This leads to significant drug interactions with other CYP450 inducers/inhibitors, giving rise to a complex therapy [105,109].

CsA and TAC are lipophilic and low solubility drugs with an oral bioavailability around 20–25%, which is variably influenced by food or fasting. The first-pass metabolism plays a key role in obtaining therapeutic blood plasma concentrations [110]. Thus, intravenous administration could improve the PK profile of the drugs. However, physiologically based PK models showed that different covariates, such as serum albumin level, hematocrit, CYP3A4 abundance, and CYP3A5 genotype status, are quantitatively responsible for PK variability, making it difficult to understand the drug disposition process [108,110]. Finally, it has been proposed that there is a requirement for pharmacogenetic screening prior to the administration of drugs in order to identify rapid or slow metabolizer patients and regulate the dosage to avoid adverse effects [111]. For instance, TAC is one of

the drugs with available guidelines to support the necessity of pharmacogenetic evaluation before and after transplantation [106]. Nevertheless, it is an expensive drug without widespread routine examination to be used in every immunosuppressed patient. It requires the consultation of health professionals to determine the appropriate genotype-based dose adjustment [112].

2.3. The Adverse Effects of CNIs

Since the 1970s, the proven efficacy of CNIs in preventing rejections has been accompanied by undesirable side effects. Among the common and prominent side effects, hypertension appears to be a relevant one, resulting in an increase in graft failure as well as recipient mortality [113]. Thus, it has been observed in patients and experimental animals that the CNIs promote vasoconstriction, which is extended even to the arteriolar level in renal afferent arteriole [114] and promoted by sympatho-excitation [115]. This results from an imbalance between the pro-contractile (endothelin and thromboxane) and pro-dilative molecules (nitric oxide, prostaglandin E2, prostacyclin) and activation of the renin-angiotensin system after the administration of CNIs [116–118]. In addition, Hoorn et al. performed a study on mice that demonstrated the positive influence of TAC on increasing the blood pressure by the activation of the renal sodium chloride cotransporter [102], which is crucial for the reabsorption of sodium at the distal nephron. This may represent an additional mechanism for the induction of hypertension during the administration of CNIs.

Nephrotoxicity is another important adverse effect of the CNIs that leads to kidney failure. The acute CNI nephrotoxicity was reported as one of the first pieces of evidence for nephrotoxicity related to CNIs, with pivotal vasoconstriction mechanisms, as mentioned above. During acute toxicity, the CsA and TAC promote thrombotic microangiopathy [119] and isometric tubular vacuolization [120], augmenting the renal risk after organ transplantation. Since renal hemodynamic alterations are crucial mechanisms in CNI-induced nephrotoxicity, the diminution in renal blood flow [121] and increased renal vascular resistance in kidneys [120] also contribute to the acute toxicity levels. On the other hand, chronic CNI-related nephrotoxicity involves the alteration of the renal structure by inducing pro-fibrotic factors such as TGFβ [122] and promoting epithelial–mesenchymal transition, a major mechanism involved in the interstitial fibrosis of the kidneys [123]. Additionally, the inflammatory vasculopathy and endothelial dysfunction promoted by CNIs contribute to tissue remodeling [124]. All these effects are reinforced by the activation of the renin-angiotensin system, resulting in acute nephrotoxicity.

The side effects of CsA and TAC pose limitations on the long-term survival of the transplant and overall prognosis of the patient receiving the organ. The non-selective inhibition induced by the CNIs on the immune system may further reduce the resistance to infections and augment the potential spread of malignant conditions [125]. In addition, the CNIs may increase the risk of cardiovascular and renal diseases through dyslipidemia and hyperglycemia [126].

The majority of the acute side effects result from the high variability in the plasma concentrations of the CNIs, depending on the dosage. This is an important issue to be considered during the determination of the appropriate dosage in patients. Drug formulation based on the technological aspects provides new perspectives in drug delivery and disposition, which could possibly optimize the stable therapeutic plasma concentration, avoid high or low exposure to the drug (wide peak-to-trough plasma fluctuations), and improve the quality of life after immunosuppressive therapy.

3. Encapsulation of the Calcineurin Inhibitors

CsA presents limitations in oral and injectable administration, where its low aqueous solubility and hydrophobic nature have encouraged the development of drug delivery systems with maximum bioavailability. Thus, natural PolyHy have been proposed as local delivery systems for the sustained release of CsA, which may be an effective way to resolve this problem [127]. Sang Shin et al. [95] proposed a formulation based on injectable hydrogels composed of calcium hyaluronic acid (HA-Ca) and sodium Alg (Alg-Na) for the sustained release system of CsA. The injectable HA-Ca-Alg hydrogels were successfully prepared at different values of gel time and volume ratios without the external

addition of calcium salts. The morphology of PolyHy was characterized and analyzed by scanning electron microscopy, which showed the existence of irregular pores. This feature offers suitable microenvironments for the transport of large-sized immunosuppressive drugs, such as CsA. The results reveal effective immunosuppressive activity for prolonged duration without any effects on blood clotting and normal cells. This study suggested the effective applicability of injectable HA-Ca-Alg PolyHy for the sustained delivery of hydrophobic immunosuppressive drugs. Nanostructured poly(2-hydroxyethyl methacrylate) hydrogels (p-HEMA) containing microemulsions have been developed for the prolonged ophthalmic administration of CsA [128], which yielded promising results. However, p-HEMA lenses cannot be worn for long duration due to their low oxygen permeability.

Studies were performed on different delivery systems for the administration of TAC, such as mPEG-poly(lactic acid) nanoparticles [83], poly (lactide-co-glycolide) microspheres [129], and triglycerol monostearate hydrogels [130]. Recently, a mixed PolyHy system based on thermosensitive polypeptides and poloxamer was proposed to develop a sustained release reservoir for the delivery of subcutaneously administered TAC in an in vivo model [131]. In addition, the rational design of two hydrogelators was reported to establish an easy method for the release of TAC that induces an immune response to overcome the organ transplant rejection in an animal model [132]. The in vitro cell experiments showed that the amount of TAC released from the hydrogel was logarithmically proportional to the number of activated T-cells, demonstrating a better cell-inhibition effect than the free TAC drug. The in vivo liver transplantation trials indicated that the smearing of PolyHy on the graft surfaces after surgery enhanced the survival rate. This could be a potential alternative to ensure therapeutic and stable concentrations of TAC during the first months of organ transplantation that exhibit critically high rates of rejection due to the inadequate plasma concentration of the immunosuppressants [112]. Another study was focused on the development of immunosuppressive prodrugs, such as TAC modified with dibenzocyclooctin (DBCO) and its conjugation to Alg PolyHy modified with an azido group via click chemistry (Figure 5) [133].

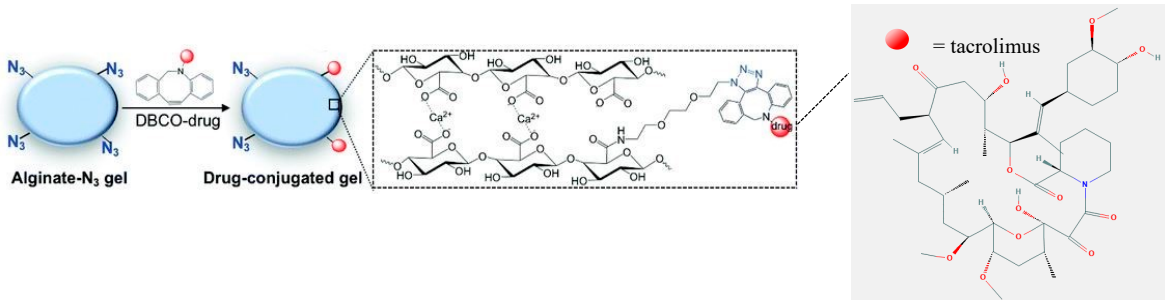

Figure 5. Schematic illustration of the binding of DBCO-drug (e.g., tacrolimus) prodrugs to the alginate-N3 gels. The schemes were extracted from reference [133]. PubChem Compound Summary for CID 445643, Tacrolimus. Retrieved 19 August 2020 from https://pubchem.ncbi.nlm.nih.gov/compound/Tacrolimus.

The Alg hydrogels were found to be non-immunogenic and showed the controlled local release of immunosuppressive drugs, such as TAC. This strategy demonstrated the intravenous injection of fresh prodrugs into the hydrogels. Therefore, there is no need to replace the implanted gels within the tissues as the drug levels drop to the subtherapeutic levels, which avoids the trauma and inflammation resulting from repeated gel injection. This alternative plays a key role in long-term adherence to the treatment. The risk factors for organ rejection include polypharmacy, adverse effects, and lack of the patient's adherence to the chronic health conditions [112]. Thus, any new technology for the reduction in drug administration and preservation of stable drug concentrations in the therapeutic range is an important advancement in therapeutics. CS, another natural polysaccharide, is also widely used and studied as a drug delivery system [134]. The chemically-modified CS compounds containing hydrophilic and hydrophobic groups (HGC) were synthesized and loaded with TAC to be

directed toward the kidney [135]. In this study, HGC-TAC showed no toxicity toward HK-2 cells and enhanced the cellular uptake. The in vivo biodistribution tests showed high renal accumulation of HGC. The in vivo TAC release profile demonstrated a higher renal concentration and lower plasma profile, indicating the suitability of HGC-TAC as a safe TAC delivery system for the kidneys while maintaining low plasma concentrations of TAC. Finally, type I collagen-based PolyHy have also been used to encapsulate primary immunosuppressive agents, such as TAC. The hydrogels were found to exhibit adequate porosity, degree of swelling, drug release, blood compatibility, and cell proliferation [136].

4. Conclusions

The PolyHy represent a special class of polymers that are extensively found in nature. These cross-linked networks possess fascinating physical and chemical properties and adopt different shapes and sizes depending on the existing environment. Compared to other polymers, these hydrogels possess the unique characteristic of water absorption and arouse great scientific interest due to their biocompatibility and versatility. As discussed in this review, the PolyHy are used as carriers for the drug delivery of certain medications that present low bioavailability and tissue distribution, variable plasma therapeutic levels, high first-pass metabolism, or even low compliance of the patient to the treatment. All of these limitations affect the pharmacological therapy and can be resolved by improving the delivery system. The CNIs, namely TAC and CsA, constitute a drug family that was proposed to be encapsulated in PolyHy for the treatment of autoimmune diseases but are more relevant for immunosuppressive treatment after solid organ transplant. The first in vitro and in vivo assays of CNIs indicated effective immunosuppressive activity and therapeutic concentrations in the target organ. The unique in vivo results were not only observed in experimental animals but also in pilot clinical organ transplant protocols, which is beneficial for the development of solid therapeutics. Despite the development of other immunosuppressants, the CNIs are potentially used for maintenance therapies after solid organ transplants due to their excellent cost-efficiency, representing the first line of drugs to prevent acute and chronic rejections [137].

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References

1. Lin, Y.-H.; Liang, H.-F.; Chung, C.-K.; Chen, M.-C.; Sung, H.-W. Physically crosslinked alginate/N,O-carboxymethyl chitosan hydrogels with calcium for oral delivery of protein drugs. *Biomaterials* **2005**, *26*, 2105–2113. [CrossRef] [PubMed]
2. Li, X.; Peng, X.; Li, R.; Zhang, Y.; Liu, Z.; Huang, Y.; Long, S.; Li, H. Multiple Hydrogen Bonds–Reinforced Hydrogels with High Strength, Shape Memory, and Adsorption Anti-Inflammatory Molecules. *Macromol. Rapid Commun.* **2020**, 2000202. [CrossRef] [PubMed]
3. Sabzi, M.; Afshari, M.J.; Babaahmadi, M.; Shafagh, N. pH-dependent swelling and antibiotic release from citric acid crosslinked poly(vinyl alcohol) (PVA)/nano silver hydrogels. *Colloids Surf. B Biointerfaces* **2020**, *188*, 110757. [CrossRef] [PubMed]
4. Liu, J.; Fan, X.; Tao, Y.; Deng, C.; Yu, K.; Zhang, W.; Deng, L.; Xiong, W. Two-Step Freezing Polymerization Method for Efficient Synthesis of High-Performance Stimuli-Responsive Hydrogels. *ACS Omega* **2020**, *5*, 5921–5930. [CrossRef]
5. Huang, J.; Jiang, X. Injectable and Degradable pH-Responsive Hydrogels via Spontaneous Amino–Yne Click Reaction. *ACS Appl. Mater. Interfaces* **2018**, *10*, 361–370. [CrossRef]

6. Rajan, R.; Pangkom, N.; Matsumura, K. Design of Stimuli-Responsive Polyampholytes and Their Transformation into Micro-Hydrogels for Drug Delivery. In *Polymers in Therapeutic Delivery*; ACS Symposium Series; American Chemical Society: Washington, DC, USA, 2020; Volume 1350, pp. 47–62. ISBN 978-0-8412-3814-5.
7. Liu, Q.; Liu, L. Novel Light-Responsive Hydrogels with Antimicrobial and Antifouling Capabilities. *Langmuir* **2019**, *35*, 1450–1457. [CrossRef]
8. Xiang, T.; Lu, T.; Zhao, W.-F.; Zhao, C.-S. Ionic-Strength Responsive Zwitterionic Copolymer Hydrogels with Tunable Swelling and Adsorption Behaviors. *Langmuir* **2019**, *35*, 1146–1155. [CrossRef]
9. Huang, K.-T.; Ishihara, K.; Huang, C.-J. Polyelectrolyte and Antipolyelectrolyte Effects for Dual Salt-Responsive Interpenetrating Network Hydrogels. *Biomacromolecules* **2019**, *20*, 3524–3534. [CrossRef] [PubMed]
10. He, H.; Xuan, X.; Zhang, C.; Song, Y.; Chen, S.; Gong, X.; Ren, B.; Zheng, J.; Wu, J. Simple Thermal Pretreatment Strategy to Tune Mechanical and Antifouling Properties of Zwitterionic Hydrogels. *Langmuir* **2019**, *35*, 1828–1836. [CrossRef]
11. GhavamiNejad, A.; Park, C.H.; Kim, C.S. In Situ Synthesis of Antimicrobial Silver Nanoparticles within Antifouling Zwitterionic Hydrogels by Catecholic Redox Chemistry for Wound Healing Application. *Biomacromolecules* **2016**, *17*, 1213–1223. [CrossRef]
12. Bakaic, E.; Smeets, N.M.B.; Badv, M.; Dodd, M.; Barrigar, O.; Siebers, E.; Lawlor, M.; Sheardown, H.; Hoare, T. Injectable and Degradable Poly(Oligoethylene glycol methacrylate) Hydrogels with Tunable Charge Densities as Adhesive Peptide-Free Cell Scaffolds. *ACS Biomater. Sci. Eng.* **2018**, *4*, 3713–3725. [CrossRef]
13. Goto, K.; Teramoto, Y. Development of chitinous nanofiber-based flexible composite hydrogels capable of cell adhesion and detachment. *Polym. J.* **2020**, *52*, 959–967. [CrossRef]
14. Singh, M.R.; Patel, S.; Singh, D. Chapter 9—Natural Polymer-based Hydrogels as Scaffolds for Tissue Engineering. In *Nanobiomaterials in Soft Tissue Engineering*; Grumezescu, A.M., Ed.; William Andrew Publishing: Norwich, NY, USA, 2016; pp. 231–260. ISBN 978-0-323-42865-1.
15. Hsu, S.; Hung, K.-C.; Chen, C.-W. Biodegradable polymer scaffolds. *J. Mater. Chem. B* **2016**, *4*, 7493–7505. [CrossRef] [PubMed]
16. Kouhi, M.; Varshosaz, J.; Hashemibeni, B.; Sarmadi, A. Injectable gellan gum/lignocellulose nanofibrils hydrogels enriched with melatonin loaded forsterite nanoparticles for cartilage tissue engineering: Fabrication, characterization and cell culture studies. *Mater. Sci. Eng. C* **2020**, *115*, 111114. [CrossRef] [PubMed]
17. Akalin, O.B.; Bayraktar, H. Alteration of cell motility dynamics through collagen fiber density in photopolymerized polyethylene glycol hydrogels. *Int. J. Biol. Macromol.* **2020**, *157*, 414–423. [CrossRef] [PubMed]
18. Arumugam, S.; Ramamoorthy, P.; Chakkarapani, L.D. Synthesis and characterizations of biocompatible polymers and carbon nanotubes-based hybrids for biomedical applications. *Int. J. Polym. Mater. Polym. Biomater.* **2020**, *69*, 786–797. [CrossRef]
19. Hernandez-Gordillo, V.; Kassis, T.; Lampejo, A.; Choi, G.; Gamboa, M.E.; Gnecco, J.S.; Brown, A.; Breault, D.T.; Carrier, R.; Griffith, L.G. Fully synthetic matrices for in vitro culture of primary human intestinal enteroids and endometrial organoids. *Biomaterials* **2020**, *254*, 120125. [CrossRef]
20. Yao, Y.; Zaw, A.M.; Anderson, D.E.J.; Hinds, M.T.; Yim, E.K.F. Fucoidan functionalization on poly(vinyl alcohol) hydrogels for improved endothelialization and hemocompatibility. *Biomaterials* **2020**, *249*, 120011. [CrossRef]
21. Van Hoorick, J.; Delaey, J.; Vercammen, H.; Van Erps, J.; Thienpont, H.; Dubruel, P.; Zakaria, N.; Koppen, C.; Van Vlierberghe, S.; Van den Bogerd, B. Designer Descemet Membranes Containing PDLLA and Functionalized Gelatins as Corneal Endothelial Scaffold. *Adv. Healthc. Mater.* **2020**, 2000760. [CrossRef]
22. Merkel, T.J.; Jones, S.W.; Herlihy, K.P.; Kersey, F.R.; Shields, A.R.; Napier, M.; Luft, J.C.; Wu, H.; Zamboni, W.C.; Wang, A.Z.; et al. Using mechanobiological mimicry of red blood cells to extend circulation times of hydrogel microparticles. *Proc. Natl. Acad. Sci. USA* **2011**, *108*, 586. [CrossRef]
23. Vakkipurath Kodakkadan, Y.N.; Idzakovicova, K.; Sepitka, J.; ten Napel, D.; Safai, E.; Cigler, P.; Štěpánek, F.; Rehor, I. Arbitrarily-shaped microgels composed of chemically unmodified biopolymers. *Biomater. Sci.* **2020**, *8*, 3044–3051. [CrossRef] [PubMed]

24. Galdioli Pellá, M.C.; Simão, A.R.; Lima-Tenório, M.K.; Tenório-Neto, E.; Scariot, D.B.; Nakamura, C.V.; Rubira, A.F. Chitosan hybrid microgels for oral drug delivery. *Carbohydr. Polym.* **2020**, *239*, 116236. [CrossRef] [PubMed]
25. Jooybar, E.; Abdekhodaie, M.J.; Karperien, M.; Mousavi, A.; Alvi, M.; Dijkstra, P.J. Developing hyaluronic acid microgels for sustained delivery of platelet lysate for tissue engineering applications. *Int. J. Biol. Macromol.* **2020**, *144*, 837–846. [CrossRef] [PubMed]
26. Rose, J.C.; Gehlen, D.B.; Haraszti, T.; Köhler, J.; Licht, C.J.; De Laporte, L. Biofunctionalized aligned microgels provide 3D cell guidance to mimic complex tissue matrices. *Biomaterials* **2018**, *163*, 128–141. [CrossRef] [PubMed]
27. Compaan, A.M.; Song, K.; Chai, W.; Huang, Y. Cross-Linkable Microgel Composite Matrix Bath for Embedded Bioprinting of Perfusable Tissue Constructs and Sculpting of Solid Objects. *ACS Appl. Mater. Interfaces* **2020**, *12*, 7855–7868. [CrossRef]
28. Ji, Y. Microgels prepared from corn starch with an improved capacity for uptake and release of lysozyme. *J. Food Eng.* **2020**, *285*, 110088. [CrossRef]
29. Feldman, D. Polymers and Polymer Nanocomposites for Cancer Therapy. *Appl. Sci.* **2019**, *9*, 3899. [CrossRef]
30. Pedrosa, S.S.; Pereira, P.; Correia, A.; Moreira, S.; Rocha, H.; Gama, F.M. Biocompatibility of a Self-Assembled Crosslinkable Hyaluronic Acid Nanogel. *Macromol. Biosci.* **2016**, *16*, 1610–1620. [CrossRef]
31. Li, D.; Gao, H.; Li, M.; Chen, G.; Guan, L.; He, M.; Tian, J.; Cao, R. Nanochitin/metal ion dual reinforcement in synthetic polyacrylamide network-based nanocomposite hydrogels. *Carbohydr. Polym.* **2020**, *236*, 116061. [CrossRef]
32. Niemczyk-Soczynska, B.; Gradys, A.; Kolbuk, D.; Krzton-Maziopa, A.; Sajkiewicz, P. Crosslinking Kinetics of Methylcellulose Aqueous Solution and Its Potential as a Scaffold for Tissue Engineering. *Polymers* **2019**, *11*, 1772. [CrossRef]
33. Cernadas, T.; Santos, M.; Gonçalves, F.A.M.M.; Alves, P.; Correia, T.R.; Correia, I.J.; Ferreira, P. Functionalized polyester-based materials as UV curable adhesives. *Eur. Polym. J.* **2019**, *120*, 109196. [CrossRef]
34. Cross, L.M.; Shah, K.; Palani, S.; Peak, C.W.; Gaharwar, A.K. Gradient nanocomposite hydrogels for interface tissue engineering. *Nanomed. Nanotechnol. Biol. Med.* **2018**, *14*, 2465–2474. [CrossRef] [PubMed]
35. Moshe, H.; Davizon, Y.; Raskin, M.M.; Sosnik, A. Novel poly(vinyl alcohol)-based amphiphilic nanogels by non-covalent boric acid crosslinking of polymeric micelles. *Biomater. Sci.* **2017**, *5*, 2295–2309. [CrossRef]
36. Huang, H.; Han, L.; Fu, X.; Wang, Y.; Yang, Z.; Pan, L.; Xu, M. Multiple Stimuli Responsive and Identifiable Zwitterionic Ionic Conductive Hydrogel for Bionic Electronic Skin. *Adv. Electron. Mater.* **2020**, *6*. [CrossRef]
37. Shriky, B.; Kelly, A.; Isreb, M.; Babenko, M.; Mahmoudi, N.; Rogers, S.; Shebanova, O.; Snow, T.; Gough, T. Pluronic F127 thermosensitive injectable smart hydrogels for controlled drug delivery system development. *J. Colloid Interface Sci.* **2020**, *565*, 119–130. [CrossRef] [PubMed]
38. Sarwar, M.S.; Huang, Q.; Ghaffar, A.; Abid, M.A.; Zafar, M.S.; Khurshid, Z.; Latif, M. A Smart Drug Delivery System Based on Biodegradable Chitosan/Poly(allylamine hydrochloride) Blend Films. *Pharmaceutics* **2020**, *12*, 131. [CrossRef]
39. Wei, H.; Liu, Z.; Zhu, H.; He, J.; Li, J. Preparation and Characterization of Thermal and pH Dual Sensitive Hydrogel Based on 1,3-Dipole Cycloaddition Reaction. *Polym. Eng. Sci.* **2020**, *60*, 917–924. [CrossRef]
40. Muhammad, G.; Haseeb, M.T.; Hussain, M.A.; Ashraf, M.U.; Farid-ul-Haq, M.; Zaman, M. Stimuli-responsive/smart tablet formulations (under simulated physiological conditions) for oral drug delivery system based on glucuronoxylan polysaccharide. *Drug Dev. Ind. Pharm.* **2020**, *46*, 122–134. [CrossRef]
41. Barkat, K.; Ahmad, M.; Minhas, M.U.; Khalid, I.; Malik, N.S. Chondroitin sulfate-based smart hydrogels for targeted delivery of oxaliplatin in colorectal cancer: Preparation, characterization and toxicity evaluation. *Polym. Bull.* **2019**. [CrossRef]
42. Huang, K.-T.; Hsieh, P.-S.; Dai, L.-G.; Huang, C.-J. Complete zwitterionic double network hydrogels with great toughness and resistance against foreign body reaction and thrombus. *J. Mater. Chem. B* **2020**, *8*, 7390–7402. [CrossRef]
43. Wang, Q.; Zhang, G.; Zhang, L.; Zhu, P. Mechanical Properties and Water Absorption of Alginate/hydroxypropyl Methyl Cellulose Blend Membranes with Semi-interpenetrating Network. *Fibers Polym.* **2020**, *21*, 1403–1410. [CrossRef]

44. Erfani, A.; Flynn, N.H.; Aichele, C.P.; Ramsey, J.D. Encapsulation and delivery of protein from within poly(sulfobetaine) hydrogel beads. *J. Appl. Polym. Sci.* **2020**, *137*. [CrossRef]
45. Boi, S.; Rouatbi, N.; Dellacasa, E.; Lisa, D.D.; Bianchini, P.; Monticelli, O.; Pastorino, L. Alginate microbeads with internal microvoids for the sustained release of drugs. *Int. J. Biol. Macromol.* **2020**, *156*, 454–461. [CrossRef] [PubMed]
46. Liu, B.; Huang, W.; Yang, G.; An, Y.; Yin, Y.; Wang, N.; Jiang, B. Preparation of gelatin/poly (gamma-glutamic acid) hydrogels with stimulated response by hot-pressing preassembly and radiation crosslinking. *Mater. Sci. Eng. C Mater. Biol. Appl.* **2020**, *116*. [CrossRef] [PubMed]
47. Moradi, S.; Barati, A.; Tonelli, A.E.; Hamedi, H. Effect of clinoptilolite on structure and drug release behavior of chitosan/thyme oil gamma-Cyclodextrininclusion compound hydrogels. *J. Appl. Polym. Sci.* **2020**. [CrossRef]
48. Khan, M.U.A.; Raza, M.A.; Razak, S.I.A.; Abdul Kadir, M.R.; Haider, A.; Shah, S.A.; Mohd Yusof, A.H.; Haider, S.; Shakir, I.; Aftab, S. Novel functional antimicrobial and biocompatible arabinoxylan/guar gum hydrogel for skin wound dressing applications. *J. Tissue Eng. Regen. Med.* **2020**. [CrossRef] [PubMed]
49. Prabhu, S.; Bubbly, S.G.; Gudennavar, S.B. Synthetic polymer hydrogels as potential tissue phantoms in radiation therapy and dosimetry. *Biomed. Phys. Eng. Express* **2020**, *6*. [CrossRef]
50. Leite Dutra, M.A.; Marques, N.N.; Fernandes, R.d.S.; Moreira de Souza Filho, M.d.S.; Balaban, R.d.C. ECO-FRIENDLY hybrid hydrogels for detection of phenolic RESIDUES in water using SERS. *Ecotoxicol. Environ. Saf.* **2020**, *200*. [CrossRef]
51. Beard, M.C.; Cobb, L.H.; Grant, C.S.; Varadarajan, A.; Henry, T.; Swanson, E.A.; Kundu, S.; Priddy, L.B. Autoclaving of Poloxamer 407 hydrogel and its use as a drug delivery vehicle. *J. Biomed. Mater. Res. Part B Appl. Biomater.* **2020**. [CrossRef]
52. Haney, B.; Werner, J.G.; Weitz, D.A.; Ramakrishnan, S. Stimuli responsive Janus microgels with convertible hydrophilicity for controlled emulsion destabilization. *Soft Matter* **2020**, *16*, 3613–3620. [CrossRef]
53. Damiati, S. In Situ Microfluidic Preparation and Solidification of Alginate Microgels. *Macromol. Res.* **2020**. [CrossRef]
54. Mahdieh, Z.; Holian, A. Electrospun fibers loaded with ball-milled poly(n-isopropylacrylamide) microgel particles for smart delivery applications. *J. Appl. Polym. Sci.* **2020**, *137*. [CrossRef]
55. Zhang, Q.; Yu, G.; Zhou, Q.; Li, J.; Feng, Y.; Wang, L.; Tang, Y.; Peng, Y. Eco-friendly interpenetrating network hydrogels integrated with natural soil colloid as a green and sustainable modifier for slow release of agrochemicals. *J. Clean. Prod.* **2020**, *269*. [CrossRef]
56. Nikfarjam, M.; Kokabi, M. Chitosan/laponite nanocomposite nanogels as a potential drug delivery system. *Polym. Bull.* **2020**. [CrossRef]
57. Horvat, S.; Yu, Y.; Boejte, S.; Tessmer, I.; Lowman, D.W.; Ma, Z.; Williams, D.L.; Beilhack, A.; Albrecht, K.; Groll, J. Engineering Nanogels for Drug Delivery to Pathogenic Fungi Aspergillus fumigatus by Tuning Polymer Amphiphilicity. *Biomacromolecules* **2020**, *21*, 3112–3121. [CrossRef]
58. Qi, X.; Su, T.; Zhang, M.; Tong, X.; Pan, W.; Zeng, Q.; Shen, J. Sustainable, flexible and biocompatible hydrogels derived from microbial polysaccharides with tailorable structures for tissue engineering. *Carbohydr. Polym.* **2020**, *237*, 116160. [CrossRef]
59. Liu, L.; Zhang, Y.; Yu, S.; Yang, Z.; He, C.; Chen, X. Dual Stimuli-Responsive Nanoparticle-Incorporated Hydrogels as an Oral Insulin Carrier for Intestine-Targeted Delivery and Enhanced Paracellular Permeation. *ACS Biomater. Sci. Eng.* **2018**, *4*, 2889–2902. [CrossRef]
60. Zhou, M.; Hu, Q.; Wang, T.; Xue, J.; Luo, Y. Alginate hydrogel beads as a carrier of low density lipoprotein/pectin nanogels for potential oral delivery applications. *Int. J. Biol. Macromol.* **2018**, *120*, 859–864. [CrossRef]
61. Gálvez-Montón, C.; Prat-Vidal, C.; Roura, S.; Soler-Botija, C.; Bayes-Genis, A. Cardiac Tissue Engineering and the Bioartificial Heart. *Rev. Española Cardiol. (Engl. Ed.)* **2013**, *66*, 391–399. [CrossRef]
62. O'Brien, F.J. Biomaterials & scaffolds for tissue engineering. *Mater. Today* **2011**, *14*, 88–95. [CrossRef]
63. Pok, S.; Myers, J.D.; Madihally, S.V.; Jacot, J.G. A multilayered scaffold of a chitosan and gelatin hydrogel supported by a PCL core for cardiac tissue engineering. *Acta Biomater.* **2013**, *9*, 5630–5642. [CrossRef] [PubMed]

64. Efraim, Y.; Sarig, H.; Cohen Anavy, N.; Sarig, U.; de Berardinis, E.; Chaw, S.-Y.; Krishnamoorthi, M.; Kalifa, J.; Bogireddi, H.; Duc, T.V.; et al. Biohybrid cardiac ECM-based hydrogels improve long term cardiac function post myocardial infarction. *Acta Biomater.* **2017**, *50*, 220–233. [CrossRef] [PubMed]
65. Verma, M.L.; Dhanya, B.S.; Sukriti; Rani, V.; Thakur, M.; Jeslin, J.; Kushwaha, R. Carbohydrate and protein based biopolymeric nanoparticles: Current status and biotechnological applications. *Int. J. Biol. Macromol.* **2020**, *154*, 390–412. [CrossRef] [PubMed]
66. Novak, U.; Bajić, M.; Kõrge, K.; Oberlintner, A.; Murn, J.; Lokar, K.; Triler, K.V.; Likozar, B. From waste/residual marine biomass to active biopolymer-based packaging film materials for food industry applications—A review. *Phys. Sci. Rev.* **2019**, *5*, 20190099. [CrossRef]
67. Martău, G.A.; Mihai, M.; Vodnar, D.C. The Use of Chitosan, Alginate, and Pectin in the Biomedical and Food Sector—Biocompatibility, Bioadhesiveness, and Biodegradability. *Polymers* **2019**, *11*, 1837. [CrossRef]
68. Tamimi, M.; Rajabi, S.; Pezeshki-Modaress, M. Cardiac ECM/chitosan/alginate ternary scaffolds for cardiac tissue engineering application. *Int. J. Biol. Macromol.* **2020**, *164*, 389–402. [CrossRef]
69. Dunn, D.A.; Hodge, A.J.; Lipke, E.A. Biomimetic materials design for cardiac tissue regeneration. *WIREs Nanomed. Nanobiotechnol.* **2014**, *6*, 15–39. [CrossRef]
70. Zhang, J.; Zhou, A.; Deng, A.; Yang, Y.; Gao, L.; Zhong, Z.; Yang, S. Pore architecture and cell viability on freeze dried 3D recombinant human collagen-peptide (RHC)–chitosan scaffolds. *Mater. Sci. Eng. C* **2015**, *49*, 174–182. [CrossRef]
71. Campos, F.; Bonhome-Espinosa, A.B.; Chato-Astrain, J.; Sánchez-Porras, D.; García-García, Ó.D.; Carmona, R.; López-López, M.T.; Alaminos, M.; Carriel, V.; Rodriguez, I.A. Evaluation of Fibrin-Agarose Tissue-Like Hydrogels Biocompatibility for Tissue Engineering Applications. *Front. Bioeng. Biotechnol.* **2020**, *8*, 596. [CrossRef] [PubMed]
72. Bonhome-Espinosa, A.B.; Campos, F.; Durand-Herrera, D.; Sánchez-López, J.D.; Schaub, S.; Durán, J.D.G.; Lopez-Lopez, M.T.; Carriel, V. In vitro characterization of a novel magnetic fibrin-agarose hydrogel for cartilage tissue engineering. *J. Mech. Behav. Biomed. Mater.* **2020**, *104*, 103619. [CrossRef]
73. Moghimiardekani, A.; Molina, B.G.; Enshaei, H.; del Valle, L.J.; Perez-Madrigal, M.M.; Estrany, F.; Aleman, C. Semi-Interpenetrated Hydrogels-Microfibers Electroactive Assemblies for Release and Real-Time Monitoring of Drugs. *Macromol. Biosci.* **2020**. [CrossRef] [PubMed]
74. Schneible, J.D.; Shi, K.; Young, A.T.; Ramesh, S.; He, N.; Dowdey, C.E.; Dubnansky, J.M.; Libya, R.L.; Gao, W.; Santiso, E.; et al. Modified gaphene oxide (GO) particles in peptide hydrogels: A hybrid system enabling scheduled delivery of synergistic combinations of chemotherapeutics. *J. Mater. Chem. B* **2020**, *8*, 3852–3868. [CrossRef] [PubMed]
75. Kurochkin, M.A.; Sindeeva, O.A.; Brodovskaya, E.P.; Gai, M.; Frueh, J.; Su, L.; Sapelkin, A.; Tuchin, V.V.; Sukhorukov, G.B. Laser-triggered drug release from polymeric 3-D micro-structured films via optical fibers. *Mater. Sci. Eng. C Mater. Biol. Appl.* **2020**, *110*. [CrossRef] [PubMed]
76. Ailincai, D.; Gavril, G.; Marin, L. Polyvinyl alcohol boric acid—A promising tool for the development of sustained release drug delivery systems. *Mater. Sci. Eng. C Mater. Biol. Appl.* **2020**, *107*. [CrossRef]
77. Caroline, S.; Lady, J.; Prasetyanto, E.A. Nanocomposite mesoporous silica-alginate hydrogels for extended delivery of antibiotics. *IOP Conf. Ser. Mater. Sci. Eng.* **2020**, *858*, 012032. [CrossRef]
78. Cinay, G.E.; Erkoc, P.; Alipour, M.; Hashimoto, Y.; Sasaki, Y.; Akiyoshi, K.; Kizilel, S. Nanogel-Integrated pH-Responsive Composite Hydrogels for Controlled Drug Delivery. *ACS Biomater. Sci. Eng.* **2017**, *3*, 370–380. [CrossRef]
79. Gálisová, A.; Jirátová, M.; Rabyk, M.; Sticová, E.; Hájek, M.; Hrubý, M.; Jirák, D. Glycogen as an advantageous polymer carrier in cancer theranostics: Straightforward in vivo evidence. *Sci. Rep.* **2020**, *10*, 10411. [CrossRef]
80. Quintero-Ortega, I.A.; Romero-Argote, F.J.; Tavares-Negrete, J.A.; Elizalde-Pena, E.A.; Carvajal Garcia, Z.Y.; Perez-Perez, C.I.; Sanchez, I.C.; Luna-Barcenas, G.; Rosillo-de la Torre, A. Synthesis and characterization of simple and binary drug delivery systems for sustainable release of ciprofloxacin. *Int. J. Polym. Mater. Polym. Biomater.* **2019**, *68*, 751–761. [CrossRef]
81. Silva, J.; Vanat, P.; Marques-da-Silva, D.; Rodrigues, J.R.; Lagoa, R. Metal alginates for polyphenol delivery systems: Studies on crosslinking ions and easy-to-use patches for release of protective flavonoids in skin. *Bioact. Mater.* **2020**, *5*, 447–457. [CrossRef]
82. Motealleh, A.; Dorri, P.; Kehr, N.S. Injectable polymer/nanomaterial composites for the fabrication of three-dimensional biomaterial scaffolds. *Biomed. Mater.* **2020**, *15*. [CrossRef]

83. Paques, J.P.; van der Linden, E.; van Rijn, C.J.M.; Sagis, L.M.C. Preparation methods of alginate nanoparticles. *Adv. Colloid Interface Sci.* **2014**, *209*, 163–171. [CrossRef] [PubMed]
84. Liu, C.; Shi, Z.; Sun, H.; Mujuni, C.J.; Zhao, L.; Wang, X.; Huang, F. Preparation and characterization of tissue-factor-loaded alginate: Toward a bioactive hemostatic material. *Carbohydr. Polym.* **2020**, 116860. [CrossRef] [PubMed]
85. Araújo, M.; Bidarra, S.J.; Alves, P.M.; Valcarcel, J.; Vázquez, J.A.; Barrias, C.C. Coumarin-grafted blue-emitting fluorescent alginate as a potentially valuable tool for biomedical applications. *J. Mater. Chem. B* **2020**, *8*, 813–825. [CrossRef] [PubMed]
86. Abbasian, M.; Massoumi, B.; Mohammad-Rezaei, R.; Samadian, H.; Jaymand, M. Scaffolding polymeric biomaterials: Are naturally occurring biological macromolecules more appropriate for tissue engineering? *Int. J. Biol. Macromol.* **2019**, *134*, 673–694. [CrossRef]
87. Jahanban-Esfahlan, R.; Derakhshankhah, H.; Haghshenas, B.; Massoumi, B.; Abbasian, M.; Jaymand, M. A bio-inspired magnetic natural hydrogel containing gelatin and alginate as a drug delivery system for cancer chemotherapy. *Int. J. Biol. Macromol.* **2020**, *156*, 438–445. [CrossRef] [PubMed]
88. Moody, C.T.; Palvai, S.; Brudno, Y. Click cross-linking improves retention and targeting of refillable alginate depots. *Acta Biomater.* **2020**, *112*, 112–121. [CrossRef] [PubMed]
89. García-Astrain, C.; Avérous, L. Synthesis and evaluation of functional alginate hydrogels based on click chemistry for drug delivery applications. *Carbohydr. Polym.* **2018**, *190*, 271–280. [CrossRef]
90. Yuan, S.; Li, X.; Shi, X.; Lu, X. Preparation of multiresponsive nanogels and their controlled release properties. *Colloid Polym. Sci.* **2019**, *297*, 613–621. [CrossRef]
91. Uyen, N.T.T.; Hamid, Z.A.A.; Thi, L.A.; Ahmad, N.B. Synthesis and characterization of curcumin loaded alginate microspheres for drug delivery. *J. Drug Deliv. Sci. Technol.* **2020**, *58*, 101796. [CrossRef]
92. Potiwiput, S.; Tan, H.; Yuan, G.; Li, S.; Zhou, T.; Li, J.; Jia, Y.; Xiong, D.; Hu, X.; Ling, Z.; et al. Dual-crosslinked alginate/carboxymethyl chitosan hydrogel containing in situ synthesized calcium phosphate particles for drug delivery application. *Mater. Chem. Phys.* **2020**, *241*, 122354. [CrossRef]
93. Singh, B.; Kumar, A. Rohit Synthesis and characterization of alginate and sterculia gum based hydrogel for brain drug delivery applications. *Int. J. Biol. Macromol.* **2020**, *148*, 248–257. [CrossRef] [PubMed]
94. Mirrahimi, M.; Beik, J.; Mirrahimi, M.; Alamzadeh, Z.; Teymouri, S.; Mahabadi, V.P.; Eslahi, N.; Tazehmahalleh, F.E.; Ghaznavi, H.; Shakeri-Zadeh, A.; et al. Triple combination of heat, drug and radiation using alginate hydrogel co-loaded with gold nanoparticles and cisplatin for locally synergistic cancer therapy. *Int. J. Biol. Macromol.* **2020**, *158*, 617–626. [CrossRef] [PubMed]
95. Kim, H.-S.; Yang, J.; Kim, K.; Shin, U.S. Biodegradable and injectable hydrogels as an immunosuppressive drug delivery system. *Mater. Sci. Eng. C* **2019**, *98*, 472–481. [CrossRef] [PubMed]
96. Eivazzadeh-Keihan, R.; Khalili, F.; Aliabadi, H.A.M.; Maleki, A.; Madanchi, H.; Ziabari, E.Z.; Bani, M.S. Alginate hydrogel-polyvinyl alcohol/silk fibroin/magnesium hydroxide nanorods: A novel scaffold with biological and antibacterial activity and improved mechanical properties. *Int. J. Biol. Macromol.* **2020**. [CrossRef] [PubMed]
97. Nishiyama, T.; Yoshizaki, N.; Kishimoto, T.; Ohsumi, K. Transient activation of calcineurin is essential to initiate embryonic development in *Xenopus laevis*. *Nature* **2007**, *449*, 341–345. [CrossRef]
98. Sommerer, C.; Giese, T. Nuclear Factor of Activated T Cells-Regulated Gene Expression as Predictive Biomarker of Personal Response to Calcineurin Inhibitors. *Ther. Drug Monit.* **2016**, *38* (Suppl. S1), S50–S56. [CrossRef] [PubMed]
99. Cure, E.; Kucuk, A.; Cumhur Cure, M. Cyclosporine therapy in cytokine storm due to coronavirus disease 2019 (COVID-19). *Rheumatol. Int.* **2020**, *40*, 1177–1179. [CrossRef]
100. Schreiber, S.L.; Crabtree, G.R. The mechanism of action of cyclosporin A and FK506. *Immunol. Today* **1992**, *13*, 136–142. [CrossRef]
101. Issa, N.; Kukla, A.; Ibrahim, H.N. Calcineurin inhibitor nephrotoxicity: A review and perspective of the evidence. *Am. J. Nephrol.* **2013**, *37*, 602–612. [CrossRef]
102. Hoorn, E.J.; Walsh, S.B.; McCormick, J.A.; Fürstenberg, A.; Yang, C.-L.; Roeschel, T.; Paliege, A.; Howie, A.J.; Conley, J.; Bachmann, S.; et al. The calcineurin inhibitor tacrolimus activates the renal sodium chloride cotransporter to cause hypertension. *Nat. Med.* **2011**, *17*, 1304–1309. [CrossRef]

103. Shah, S.R.; Altaf, A.; Arshad, M.H.; Mari, A.; Noorani, S.; Saeed, E.; Mevawalla, A.A.; Haq, Z.U.; Faquih, M.E. Use of Cyclosporine Therapy in Steroid Resistant Nephrotic Syndrome (SRNS): A Review. *Glob. J. Health Sci.* **2015**, *8*, 136–141. [CrossRef] [PubMed]
104. Masuda, S.; Inui, K. An up-date review on individualized dosage adjustment of calcineurin inhibitors in organ transplant patients. *Pharmacol. Ther.* **2006**, *112*, 184–198. [CrossRef] [PubMed]
105. Berends, S.E.; Strik, A.S.; Löwenberg, M.; D'Haens, G.R.; Mathôt, R.A.A. Clinical Pharmacokinetic and Pharmacodynamic Considerations in the Treatment of Ulcerative Colitis. *Clin. Pharmacokinet.* **2019**, *58*, 15–37. [CrossRef] [PubMed]
106. Birdwell, K.A.; Decker, B.; Barbarino, J.M.; Peterson, J.F.; Stein, C.M.; Sadee, W.; Wang, D.; Vinks, A.A.; He, Y.; Swen, J.J.; et al. Clinical Pharmacogenetics Implementation Consortium (CPIC) Guidelines for CYP3A5 Genotype and Tacrolimus Dosing. *Clin. Pharmacol. Ther.* **2015**, *98*, 19–24. [CrossRef] [PubMed]
107. Moes, D.J.A.R.; Swen, J.J.; den Hartigh, J.; van der Straaten, T.; van der Heide, J.J.H.; Sanders, J.S.; Bemelman, F.J.; de Fijter, J.W.; Guchelaar, H.J. Effect of CYP3A4*22, CYP3A5*3, and CYP3A Combined Genotypes on Cyclosporine, Everolimus, and Tacrolimus Pharmacokinetics in Renal Transplantation. *CPT Pharmacomet. Syst. Pharmacol.* **2014**, *3*, e100. [CrossRef] [PubMed]
108. Vanhove, T.; Annaert, P.; Kuypers, D.R.J. Clinical determinants of calcineurin inhibitor disposition: A mechanistic review. *Drug Metab. Rev.* **2016**, *48*, 88–112. [CrossRef]
109. Lange, N.W.; Salerno, D.M.; Berger, K.; Tsapepas, D.S. Using known drug interactions to manage supratherapeutic calcineurin inhibitor concentrations. *Clin. Transplant.* **2017**, *31*. [CrossRef]
110. Emoto, C.; Johnson, T.N.; Hahn, D.; Christians, U.; Alloway, R.R.; Vinks, A.A.; Fukuda, T. A Theoretical Physiologically-Based Pharmacokinetic Approach to Ascertain Covariates Explaining the Large Interpatient Variability in Tacrolimus Disposition. *CPT Pharmacomet. Syst. Pharmacol.* **2019**, *8*, 273–284. [CrossRef]
111. Hesselink, D.A.; Bouamar, R.; van Gelder, T. The Pharmacogenetics of Calcineurin Inhibitor–Related Nephrotoxicity. *Ther. Drug Monit.* **2010**, *32*, 387–393. [CrossRef]
112. Ettenger, R.; Albrecht, R.; Alloway, R.; Belen, O.; Cavaillé-Coll, M.W.; Chisholm-Burns, M.A.; Dew, M.A.; Fitzsimmons, W.E.; Nickerson, P.; Thompson, G.; et al. Meeting report: FDA public meeting on patient-focused drug development and medication adherence in solid organ transplant patients. *Am. J. Transplant.* **2018**, *18*, 564–573. [CrossRef]
113. Kasiske, B.L.; Anjum, S.; Shah, R.; Skogen, J.; Kandaswamy, C.; Danielson, B.; O'Shaughnessy, E.A.; Dahl, D.C.; Silkensen, J.R.; Sahadevan, M.; et al. Hypertension after kidney transplantation. *Am. J. Kidney Dis.* **2004**, *43*, 1071–1081. [CrossRef] [PubMed]
114. English, J.; Evan, A.; Houghton, D.C.; Bennett, W.M. Cyclosporine-induced acute renal dysfunction in the rat. Evidence of arteriolar vasoconstriction with preservation of tubular function. *Transplantation* **1987**, *44*, 135–141. [CrossRef] [PubMed]
115. Schlaich, M.P.; Grassi, G. Sympathoexcitation in calcineurin inhibitor-induced hypertension: Villain or innocent bystander? *J. Hypertens.* **2010**, *28*, 1809–1810. [CrossRef]
116. Forslund, T.; Hannonen, P.; Reitamo, S.; Fyhrquist, F. Hypertension in cyclosporin A-treated patients is independent of circulating endothelin levels. *J. Intern. Med.* **1995**, *238*, 71–75. [CrossRef] [PubMed]
117. Roullet, J.B.; Xue, H.; McCarron, D.A.; Holcomb, S.; Bennett, W.M. Vascular mechanisms of cyclosporin-induced hypertension in the rat. *J. Clin. Investig.* **1994**, *93*, 2244–2250. [CrossRef]
118. Kurtz, A.; Della Bruna, R.; Kühn, K. Cyclosporine A enhances renin secretion and production in isolated juxtaglomerular cells. *Kidney Int.* **1988**, *33*, 947–953. [CrossRef]
119. García-Martín, P.; Alarcón-Payer, C.; López-Fernández, E.; Moratalla, L.; Romero, A.; Sainz, J.; Ríos, R.; Jurado, M. Transplantation-Associated Thrombotic Microangiopathy in Patients Treated with Sirolimus and Cyclosporine as Salvage Therapy for Graft-versus-Host Disease. *Ann. Pharmacother.* **2015**, *49*, 986–994. [CrossRef]
120. Amador, C.A.; Bertocchio, J.-P.; Andre-Gregoire, G.; Placier, S.; Duong Van Huyen, J.-P.; El Moghrabi, S.; Berger, S.; Warnock, D.G.; Chatziantoniou, C.; Jaffe, I.Z.; et al. Deletion of mineralocorticoid receptors in smooth muscle cells blunts renal vascular resistance following acute cyclosporine administration. *Kidney Int.* **2016**, *89*, 354–362. [CrossRef]
121. Kihm, L.P.; Blume, C.; Seckinger, J.; Hankel, V.; Stoffler, D.; Morath, C.; Zeier, M.; Schwenger, V. Acute effects of calcineurin inhibitors on kidney allograft microperfusion visualized by contrast-enhanced sonography. *Transplantation* **2012**, *93*, 1125–1129. [CrossRef] [PubMed]

122. Maluccio, M.; Sharma, V.; Lagman, M.; Vyas, S.; Yang, H.; Li, B.; Suthanthiran, M. Tacrolimus enhances transforming growth factor-beta1 expression and promotes tumor progression. *Transplantation* **2003**, *76*, 597–602. [CrossRef]
123. Woda, C.B.; Bruneau, S.; Mak, A.L.; Haskova, Z.; Liu, K.; Ghosh, C.C.; Briscoe, D.M. Calcineurin inhibitors augment endothelial-to-mesenchymal transition by enhancing proliferation in association with cytokine-mediated activation. *Biochem. Biophys. Res. Commun.* **2019**, *519*, 667–673. [CrossRef] [PubMed]
124. Rodrigues-Diez, R.; González-Guerrero, C.; Ocaña-Salceda, C.; Rodrigues-Diez, R.R.; Egido, J.; Ortiz, A.; Ruiz-Ortega, M.; Ramos, A.M. Calcineurin inhibitors cyclosporine A and tacrolimus induce vascular inflammation and endothelial activation through TLR4 signaling. *Sci. Rep.* **2016**, *6*, 27915. [CrossRef] [PubMed]
125. Söderlund, C.; Rådegran, G. Immunosuppressive therapies after heart transplantation—The balance between under- and over-immunosuppression. *Transplant. Rev. (Orlando)* **2015**, *29*, 181–189. [CrossRef] [PubMed]
126. Markell, M.S.; Armenti, V.; Danovitch, G.; Sumrani, N. Hyperlipidemia and glucose intolerance in the post-renal transplant patient. *J. Am. Soc. Nephrol.* **1994**, *4*, S37–S47. [PubMed]
127. Tuladhar, A.; Morshead, C.M.; Shoichet, M.S. Circumventing the blood–brain barrier: Local delivery of cyclosporin A stimulates stem cells in stroke-injured rat brain. *J. Control. Release* **2015**, *215*, 1–11. [CrossRef] [PubMed]
128. Kapoor, Y.; Chauhan, A. Ophthalmic delivery of Cyclosporine A from Brij-97 microemulsion and surfactant-laden p-HEMA hydrogels. *Int. J. Pharm.* **2008**, *361*, 222–229. [CrossRef]
129. Tajdaran, K.; Shoichet, M.S.; Gordon, T.; Borschel, G.H. A novel polymeric drug delivery system for localized and sustained release of tacrolimus (FK506). *Biotechnol. Bioeng.* **2015**, *112*, 1948–1953. [CrossRef]
130. Gajanayake, T.; Olariu, R.; Leclère, F.M.; Dhayani, A.; Yang, Z.; Bongoni, A.K.; Banz, Y.; Constantinescu, M.A.; Karp, J.M.; Vemula, P.K.; et al. A single localized dose of enzyme-responsive hydrogel improves long-term survival of a vascularized composite allograft. *Sci. Transl. Med.* **2014**, *6*, 249ra110. [CrossRef]
131. Lin, H.-C.; Anggelia, M.R.; Cheng, C.-C.; Ku, K.-L.; Cheng, H.-Y.; Wen, C.-J.; Wang, A.Y.L.; Lin, C.-H.; Chu, I.-M. A Mixed Thermosensitive Hydrogel System for Sustained Delivery of Tacrolimus for Immunosuppressive Therapy. *Pharmaceutics* **2019**, *11*, 413. [CrossRef]
132. Wu, J.; Zheng, Z.; Chong, Y.; Li, X.; Pu, L.; Tang, Q.; Yang, L.; Wang, X.; Wang, F.; Liang, G. Immune Responsive Release of Tacrolimus to Overcome Organ Transplant Rejection. *Adv. Mater.* **2018**, *30*, 1805018. [CrossRef]
133. Wang, H.; Sobral, M.C.; Snyder, T.; Brudno, Y.; Gorantla, V.S.; Mooney, D.J. Clickable, acid labile immunosuppressive prodrugs for in vivo targeting. *Biomater. Sci.* **2020**, *8*, 266–277. [CrossRef] [PubMed]
134. Wang, J.-L.; Gan, Y.-J.; Iqbal, S.; Jiang, W.; Yuan, Y.-Y.; Wang, J. Delivery of tacrolimus with cationic lipid-assisted nanoparticles for ulcerative colitis therapy. *Biomater. Sci.* **2018**, *6*, 1916–1922. [CrossRef] [PubMed]
135. Kim, C.S.; Mathew, A.P.; Uthaman, S.; Moon, M.J.; Bae, E.H.; Kim, S.W.; Park, I.-K. Glycol chitosan-based renal docking biopolymeric nanomicelles for site-specific delivery of the immunosuppressant. *Carbohydr. Polym.* **2020**, *241*, 116255. [CrossRef] [PubMed]
136. Nabavi, M.H.; Salehi, M.; Ehterami, A.; Bastami, F.; Semyari, H.; Tehranchi, M.; Nabavi, M.A.; Semyari, H. A collagen-based hydrogel containing tacrolimus for bone tissue engineering. *Drug Deliv. Transl. Res.* **2020**, *10*, 108–121. [CrossRef] [PubMed]
137. Kaplan, B.; Barr, M.L.; O'Grady, J.; Berezan, D.; Hughes, G.; Becker, B.; Blumberg, E.; Bradley, J.A.; Brennan, D.; Briscoe, D.M.; et al. *KDIGO Clinical Practice Guideline for the Care of Kidney Transplant Recipients*; Wiley Periodicals: Bethesda, MD, USA, 2009.