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Review Article

## Transdermal patches fabricated from hyaluronic acid for the enhanced skin penetration of therapeutic entities

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### ABSTRACT

Skin is an attractive route for drug delivery. However poor permeation of drugs across the skin due to the presence of extremely ordered architecture of outermost layer of skin, led to several investigation to improve the permeability of drugs. Polysaccharides remain widely studied biomaterial for the sustained delivery of drug molecules across the skin. The advance of hyaluronic acid (HA) chemistry with multiple benefits has improved the attention of research groups for its application in the skin transportation of drug molecules. Beginning from the advantages of transdermal route, the present review details the application of HA in transdermal drug delivery. In the last few decades, substantial investigation in the domain has improved the requirement for an outline of all the developments, which is depicted in the review. The review also presented different types of HA based transdermal devices such as transferosomes, nanoemulsions, microneedle etc and their potential to improve the transdermal drug delivery. Furthermore the application of HA through chemical modification as a potential transdermal device is also presented.

**Keywords:** Hyaluronic acid, transdermal drug delivery, microneedles, nanoemulsion, hydrogel

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### INTRODUCTION

Skin, the largest and easily approachable organ of the human body, is an excellent gateway for the delivery of therapeutic molecules. transdermal delivery offers avoidance of first pass hepatic metabolism, possibility of self-administration, painless drug administration, easy termination of therapy and improved patient's compliances<sup>1</sup>. However the unique architecture of skin furnishes a barrier to the drug molecules, limiting the practical application of transdermal route<sup>2</sup>. A research group in 1924 reported that the outermost layer of skin called stratum corneum (SC), of approximately 15  $\mu\text{m}$  thickness is the principal barrier to the drug transport<sup>3</sup>. Further investigation disclosed that certain drug molecules possessed remarkable skin penetration, which led to the development of transdermal devices<sup>4</sup>. For transdermal therapy to be potential, the drug molecules have to reach the vasculature in desired amounts to generate a therapeutic effect. Presently several transdermal devices are employing to overcome skin barrier function. This article discussed the application of HA in the development of transdermal drug delivery system.

HA is a linear biodegradable polymer discovered in 1934 by Karl Meyer and John Palmer in the vitreous body of the cattle

eye. It is an anionic, unbranched, non-sulfated glycosamino glycan consisted of alternating units of D-glucuronic acid and N-acetyl-D-glucosamine linked by  $\beta(1,4)$  and  $\beta(1,3)$  glycosidic bonds. HA naturally occurs in the extracellular matrix of connective tissues, dermal layer of skin, vitreous humor of eye and synovial fluids of joints<sup>5</sup>. The physiological function of HA includes tissue repairing, wound healing, maintains tissue hydrodynamics, acts as a lubricant in muscular connective tissues etc.

HA's immunoneutrality brands it as a fascinating building constituent for biomaterials to be utilized for biomedical applications including drug delivery, tissue engineering etc. The important physicochemical characteristics that grant to its success in ophthalmologic surgery is the semiflexible behavior of the biodegradable HA chains. The shear thinning property of HA solutions ultimately results in the reduction of viscosity as the molecules align in the streamlines of flow. This response makes HA simple to handle while pushing them through medical needles<sup>5</sup>. HA can be chemically conjugated to drug carriers or directly to therapeutic molecules. The carboxylic and primary hydroxyl moieties present in HA furnish favorable sites for chemical modifications. The primary hydroxyl moieties allows esterification and

etherification while carboxylic moieties permits ester and amide bond formation<sup>6</sup>. The most remarkable benefit is the facile association of therapeutic molecules with the biopolymer which avoids problems associated with solubility. Furthermore HA enhances half-life of drugs in blood plasma and possess tumor targeting ability<sup>6</sup>.

HA is an fascinating platform for transdermal drug delivery. The transdermal application of HA moisturize the surface of skin which assists the transdermal absorption of therapeutic molecules and can promote the retention of these molecules in the comparatively hydrated epidermal layers<sup>7</sup>. Yang et al detailed that HA possess the capacity to disrupt and penetrates skin layers and cotransport the therapeutic molecules across the skin<sup>8</sup>. The skin penetration enhancement of HA is due to the synergistic effect of co-transport mechanism, hydration of skin and its interaction with keratin architecture of skin<sup>7</sup>. However for the development of effective transdermal drug delivery system based on HA, several chemical and physical modification are required. The details of these modification and the capacity of hyaluronic acid to transport several drugmolecules are elucidated below.

### HA based transdermal drug delivery systems

El Refaie et al prepared a transdermal system, gel-core hyalurosomes to improve the skin transportation of curcumin for wound healing<sup>9</sup>. The curcumin loaded gel-core hyalurosomes was fabricated using thin film evaporation strategy. The dry film obtained from Lipoid S100 and Tween 80 was hydrated with HA followed by the loading of curcumin. *In vivo* study disclosed that the application of the developed device in rat burn-wounds resulted in its swift healing ascribed to the anti-inflammatory effect of curcumin, in addition to the effect of HA. The device could enhance the skin transportation of curcumin, securing it against metabolism, hence improving the bioavailability.

Lim et al prepared a hydrogel nanoparticle by the crosslinking reaction of HA and polyethylene glycol<sup>10</sup>. The research group identified that the device displayed different capacity to permeate into skin depending on the nature of dispersion medium. Compared to hydrogel dispersed in an oil-in-water emulsion, that dispersed in oil or an oil composition showed better results. They reported that if the hydrogel carried drug molecules, it might release the drug by expanding its volume by the uptake of water present in the surface of skin. The enhanced water uptake is ascribed to the feature of HA.

Wei et al employed HA as matrix of nanocrystal based hydrogel and evaluated its capacity to enhance the transdermal delivery of Baicalin<sup>11</sup>. The hydrogel comprising of 1.0 % HA displayed that the highest gelation ability and thinning shear rheological properties. *In vitro* permeation test revealed a significant improvement in the permeation of Baicalin from HA based hydrogel compared to that of coarse gel. Cilurzo et al studied the permeation behavior of HA and their sulfates across the skin<sup>12</sup>. The permeation test revealed that the sulfated HA permeated efficiently than corresponding HA. However the skin permeability was significantly reduced when the molecular mass of the polymer was increased. They also reported that HA possessed remarkable affinity for corneocytes and hence permeate chiefly through the transcellular route. The molecular dynamics analysis indicated that the increase in permeability of the studied polysaccharide was ascribed to their ability to assume flexible and extended conformations.

Franze et al fabricated a transdermal system based on HA decorated liposomes for the delivery of nifedipine<sup>13</sup>. They prepared liposome formulations by thin film hydration

method using egg phosphatidylcholine, Tween 80 and nifedipine. The obtained lipid film was hydrated using HA-1,2-dipalmitoyl-sn-glycero-3-phosphoethanolamine conjugate. The outcomes disclosed that the presence of HA in the device played a remarkable role in the potential penetration of liposomes. But the presence of HA notably influenced the flexibility of the device ascribed to the enhanced lipid chain packing. However the vesicle fluidity play a key role in enhancing the skin permeation which counters balanced the stiffening effect. When HA is incorporated to vesicle in co-administration, HA penetrate first into the skin layers, filling the holes for the diffusion of liposomes.

Gao et al enhanced the transdermal delivery of 10,11-Methylenedioxycompotothecin using HA nanoemulsions<sup>14</sup>. Glycerol  $\alpha$ -monostearate was conjugated to HA and the HA nanoemulsion with hydrophobic interior and hydrophilic interfaces was obtained through oil/water/surfactant emulsifying system and solvent evaporation. The device displayed an excellent entrapment efficiency of about 81.81 %. The fluorescence microscope images confirmed that HA nanoemulsion efficiently permeate through the epidermis and then into the dermal region. *In vitro* skin permeation test revealed that compared to control, HA nanoemulsion significantly improved the transdermal delivery of 10,11-Methylenedioxycompotothecin. The increased permeability of HA nanoemulsion is ascribed to the potential of HA to enhance skin penetration via transdermal hydration gradient and from high lipid hydration energy. The article reported that the developed device has the capacity to be employed for keloid therapy.

Kong et al studied the stability of HA based nanoemulsion and its capacity as transdermal device<sup>15</sup>. HA was modified with glycerol- $\alpha$ -monostearate and the nanoemulsion was prepared by oil/water/surfactant emulsifying system and solvent evaporation. The article reported that the stability of nanoemulsion was increased in presence of cross-linking agent, controlling the degree of substitution and modification of HA. The electrostatic, hydrophobic and steric effect involved a crucial part in the nanoemulsion stability. Furthermore, surfactants are main modulators to stabilize the colloidal interface. The device was also shown to efficiently encapsulate lipophilic molecules. In another article Kong et al reported the potential of the oil/water HA nanoemulsion to transport  $\alpha$ -tocopherol across the skin<sup>16</sup>. *In vitro* skin permeation test revealed that the developed nanoemulsion efficiently penetrate into deeper skin layers without any aid of penetration enhancers. Histological examination confirmed the security of the patch and they stated that the device enhanced the skin permeation without causing any skin irritation

A nanographene oxide-HA conjugate was developed by Jung et al for the photothermal ablation therapy of melanoma skin cancer using near infrared laser<sup>17</sup>. *In vitro* bioimaging and confocal imaging undoubtedly revealed the significant transdermal delivery of the prepared device to tumour cells, attributed to the over expressed HA receptors and comparatively leaky structures around tumor cells, permitting the improved penetration and detention of nanoparticles. The application of near infrared radiation produced total ablation of cancer cells without any recurrence of tumor genesis. The antitumor effect of the device was verified by histological assay, ELISA for caspase-3 activity and immune histochemical TUNEL assay.

Kim et al developed a potential transdermal vaccination by using two essential innovations: the utilization of HA as carrier for vaccine and non-ablative laser adjuvants<sup>18</sup>. Fluorescence assay and intravital microscopy confirmed that

HA conjugated ovalbumin vaccine patch could efficiently permeate into deeper layers of porcine and murine skins. The transdermal application of device remarkably improved the mucosal and humoral antibodies. Significant sturdy immunization was obtained with less doses of HA complex when pre-treated the skin using non-ablative laser beams. All the test outcomes suggested the capacity of the developed device for transdermal vaccination.

Kong et al prepared biocompatible doxorubicin encapsulated HA based transferosome for tumor metastasis therapy <sup>19</sup>. Amphiphilic HA obtained by the modification of HA with glycerol- $\alpha$ -monostearate was assembled on the transferosome. The *in vitro* test showed that the skin penetration of the prepared device was three times more than the solution. Furthermore, *in vivo* test revealed that the device led to remarkably greater accumulation in lymph nodes. The prepared transferosome significantly improved the endocytosis of breast tumor cells, producing nine times greater cellular uptake compared to pristine transferosome. Kong et al fabricated HA based niosome for transdermal tumor therapy <sup>20</sup>. HA was modified with monostearin and the resulting amphiphilic HA was self-assembled on the surface of niosome. The developed device had around 40 nm size, excellent stability and enhanced drug encapsulation efficiency. *In vivo* skin penetration test revealed that the nanocarrier diffused into deeper dermal layer after 8 h of application. HA accumulated around hair follicles and even penetrated throughout the aqueous surrounding of epidermis and dermis. The device displayed more endocytosis to mouse breast tumor cell than the control (Chitosan). The investigation of histological section proved the potential and security of skin penetration.

Kim et al developed a temperature sensitive transdermal device based on HA <sup>21</sup>. They synthesized double cross-linked interpenetrating polymer network hydrogels consisted of HA and poly(N-isopropyl acrylamide) and evaluated their capacity to deliver luteolin across the skin for the treatment of psoriasis. The hydrogel synthesized from 3 % crosslinker displayed the highest stability. The drug release was found to be maximum at 25 °C and pH 5.5. The release of luteolin was induced by non-Fickian mechanism at conditions similar to psoriasis skin. The test indicated that gradually released drug may assist alleviate psoriasis by inhibiting hyperproliferation of keratinocyte.

Manca et al prepared curcumin encapsulated nanovesicle using sodium hyaluronate <sup>22</sup>. They studied the *in vitro* and *in vivo* permeation capacity of the prepared nanovesicles and compared to those of liposomes. TEM and XRD data displayed that the developed nanocarrier were spherical with 112-220 nm size. Skin permeation test displayed an enhanced capacity of the nanovesicle to deposit drug molecules in a much faster way. Furthermore the nanovesicles were identified as biocompatible and safeguard human keratinocytes from the damage caused by oxidative stress and facilitated tissue remodeling. Finally, *in vitro* studies proved the capacity of the device to counteract 12-6-tetradecanoilphorhol created injuries and inflammation, reducing oedema development and furnishing skin re-epithelization.

Martins et al developed solid-in-oil nanodispersion from HA and bovine serum albumin and evaluated its potential to be employed as transdermal device <sup>23</sup>. The capacity of this nanodispersion to penetrate into deeper skin layers were confirmed by confocal and fluorescence microscopy. The article reported that the developed bovine serum albumin/HA disintegrated when it passes the SC. Noteworthy, the dispersion was found to be non-toxic in fibroblast and keratinocyte cells. The molecular weight of HA was too lower

and hence less concentrated when compared with bovine serum albumin. This assisted HA to pass into deeper layers of skin. Therefore when the nanodispersion penetrates into skin layers bovine serum albumin could be hydrolyzed by proteases residing in the medium, which makes HA remarkably available and penetrate deeply. All these factors enhanced its availability and activity to act on skin targets.

Wang et al prepared transdermal device based on HA encapsulated CuS nanoparticles. The transdermal permeation potential of the device was studied in details. The photothermal effect of the device at different concentration of HA-CuS was measured. The outcomes displayed that the device could potentially convert NIR irradiation to heat. In addition, *in vivo* experiment of laser activation of the device was carried out and the outcomes revealed that the device possessed excellent *in vivo* laser activation potential. Histological assay displayed that SC were disrupted by the application of device and exposed with 0.24 W/cm<sup>2</sup> NIR irradiation. They further studied the capacity of the device to transport insulin across the skin and found that the blood glucose level of the treated mice was substantially reduced after the application of the device and irradiated with NIR.

Smejkalov et al fabricated polymeric micelles which provided an auspicious method for the transdermal delivery of drug molecules <sup>24</sup>. The polymeric micelle was prepared from oleic acid and HA. Permeation test revealed that the HA based micelle deposited three times and six times more drug, respectively in the epidermis and dermis, compared to that of non-polymeric micelle solution. The drug molecule in epidermis and dermis was visualized using confocal imaging. The articles detailed that the chief pathway for the permeation of drug was transcellular penetration.

Wittig et al reported that HA was greatly compatible with biomolecules and can enhance the drug absorption <sup>7</sup>. They studied the effect of HA hydrogel with variable molecular weight on transdermal permeation of bovine serum albumin. In addition, the research group proved the skin-HA interaction using fluorescence life time imaging microscopy and FTIR spectroscopy. In the tape-stripped skin without any barrier, only bovine serum albumin permeated into dermal skin layers. The permeation was limited to the epidermis when the skin was applied with both bovine serum albumin and HA. In the case of normal skin, an improvement in permeation into the epidermis was identified by the application of low molecular weight HA. A close HA- bovine serum albumin was suggested by fluorescence energy transfer analysis. Furthermore FTIR studies displayed an inter-conversion of keratin from  $\alpha$ -helix to  $\beta$ -sheet. This improved the hydration of skin and interaction of lipids with HA produced a remarkable disruption in skin layers. Finally they concluded that HA behaved as a penetration enhancer for the transport of biomacromolecules in normal skin, chiefly mediated by the synergistic effect of co-transport, disruption of SC and improved hydration of skin.

Berko and coworkers prepared cross linked HA system using diamine solution via carbodiimide technique and compared its skin penetration to that of linear HA <sup>25</sup>. The tests results revealed that compared to linear HA, crosslinking of HA improved the hydration in deeper skin layers and enhanced its permeation through human epidermis. Furthermore, the dynamic rheological measurement revealed that the crosslinking changed the specific viscoelastic behavior of HA. Transepidermal water loss test showed that no skin irritation was observed after the application of crosslinked HA system. The investigation concluded that cross-linked HA could be used for the fabrication of transdermal drug delivery system with enhanced skin permeability.



The research group of Kong developed a transdermal device using HA and hydroxyethyl cellulose and used for the skin transportation of isoliquiritigenin <sup>26</sup>. HA and hydroxyethyl cellulose was crosslinked using divinyl sulfone. The developed device significantly enhanced the skin permeability of isoliquiritigenin, ascribed to the improved water retention capacity of the HA based system, which reduced the skin barrier function through hydration of skin. The fabricated HA-hydroxyethyl cellulose system displayed stable viscoelastic properties and optimal adhesiveness.

Xie et al prepared HA based ethosome and encapsulated with rhodamine B to evaluate the skin permeation <sup>27</sup>. The developed transdermal device was spherical and displayed excellent dispersion and stability. Permeation test showed that HA containing ethosome displayed greater permeation compared to that of ethosome without HA. Fluorescence microscopic investigation indicated that the device permeated into dermal layer of skin. The enhancement effect shown by HA loaded ethosome was ascribed to its smaller size, hydration of HA and significant targeting to skin and skin appendages of the liposomal carriers. Furthermore the device was proven to be non-cytotoxic and can provide a swift and safer transdermal drug delivery system.

Yang et al develop a receptor mediated transdermal system based on HA-human growth hormone conjugate <sup>28</sup>. The conjugate was prepared by the reaction of aldehyde modified HA and the amine group of human growth hormone. Fluorescence microscopy markedly indicated the significantly improved permeation of the device through mice skin. Pharmacokinetic assay suggested that the device was permeated and reached the blood circulation by the receptor mediated transdermal delivery. The results proved the ability of the system to deliver protein drugs across the skin.

Yue et al developed a HA based transdermal device for bupivacaine delivery. They conjugated HA and linolenic acid with propylene glycol <sup>29</sup>. The device displayed small particle size of 150 nm, excellent stability and drug encapsulation efficiency of about 90 %. Compared with free bupivacaine, the device showed 2.5 fold enhancements in skin permeation and prolonged antinociceptive effect.

### HA based transdermal microneedles patches

Choi et al fabricated microneedles comprising cross-linked HA by micromolding strategy <sup>30</sup>. HA was cross-linked using 1,4-butanediol diglycidylether. The study on swelling feature indicated that the swelling ratio of the cross-linked HA was significantly greater than that of pure HA. The developed microneedles have diameter of 90  $\mu\text{m}$  and 10  $\mu\text{m}$ , respectively at the base and tip. The permeation test revealed that a delay in HA degradation and sustained release profile can be obtained from the developed microneedle. Confocal microscopy confirmed the ability of the microneedle to deliver fluorescein into the skin.

Kim et al developed a dissolving HA based MN device encapsulated with ascorbic acid-2-glucoside <sup>31</sup>. The device was sterilized by gamma ray and electron beam. The microneedle architectures maintained their morphology after the application of electron beam and gamma rays. Nevertheless, the irradiation with gamma rays remarkably degraded the loaded ascorbic acid-2-glucoside while an electron beam of 40 kGy did not affect the release portrait and dissolution rate of microneedles.

Matsuo et al detailed a vaccine therapy for Alzheimer's diseases by employing microneedles fabricated from HA <sup>32</sup>. The developed device potentially induced anti-A $\beta$ 1-42 immuno response. Furthermore, the device has several

benefits as it was simple, ease of application and minimally invasive vaccination strategy. Finally they proposed that certain modifications should be required for some downsides associated with the device. Wu et al fabricated a microneedle device based on HA loaded with sumatriptan succinate for migraine treatment <sup>33</sup>. The devices conserved their skin penetration potential for 30 minutes after the application. *In vitro* test demonstrated a quick release profile of sumatriptan succinate from the device. Optical coherence tomography displayed that the device was efficiently penetrated the skin with no cracking or bending. The device was entirely dissolved in 60 minutes. The application of device resulted in a dose-dependent plasma sumatriptan succinate concentration. The delivery of sumatriptan succinate was similar to that from subcutaneous injection and high bioavailability which was significantly greater than that created by oral administration.

Liu et al examined the features of extending-4 tip-loaded microneedles prepared from HA and further compared their potential with subcutaneous injection in diabetic rats <sup>34</sup>. The release of encapsulated fluorescence isothiocyanate labeled dextran from the device was very quick, especially in the first 30 s and within 600 s, most of the loaded molecules were released. Furthermore, glucose tolerance was improved and the release of insulin was improved when the device was applied. They reported that these outcomes were comparable with that of subcutaneous injection. The plasma concentration portraits suggested that the device can be employed as an alternative system for the treatment of type 2 diabetics. In another article Liu et al described the fabrication of insulin loaded HA microneedles and the features of the device such as stability, hygroscopy and drug release profile were examined <sup>35</sup>. The developed microneedles possessed 800  $\mu\text{m}$  length with 160 and 40  $\mu\text{m}$  base and tip diameter, respectively. When the insulin loaded device was stored for 30 days at different temperatures, no significant loss in insulin was observed. More than 90 % of the loaded insulin was remained in the microneedles at all the studied temperature, which suggested the stability of the device. Furthermore, a remarkable enhancement in transepidermal water loss was identified after the insertion of microneedles and the damage produced by the device was reversible. *In vitro* permeation test indicated that insulin was swiftly released from the device. *In vivo* test displayed that a dose-dependent hypoglycemic effect and the transdermal delivery of insulin was identified after the insertion of patches.

Yu et al prepared microneedle based on alginate and HA for transdermal insulin delivery <sup>36</sup>. The device demonstrated superior mechanical strength to pierce deep into the skin and excellent degradability to deliver the encapsulated insulin. Confocal images confirmed that insulin could moderately diffuse to deeper skin layers. The loaded insulin in the device could maintain the high pharmacological activity, confirming a sustained hypoglycemic effect. The relative pharmacologic availability of insulin from the device was 90.5 $\pm$ 6.8 %. while relative bioavailability was 92.9 $\pm$ 7 %.

### CONCLUSIONS

From the plethora of investigation involving HA in the last few years, it is obvious that HA have the capacity to potentially enhance the transdermal delivery of drug molecules. HA based hydrogel and nanoemulsion furnished potential value as transdermal device in addition to niosomes and transferosomes. HA based microneedles have also displayed appreciable guarantee in disturbing lipid layers of skin so as to improve the skin transportation. However little is studied about the mechanism in which HA enhances the skin permeation. Although HA promises to revolutionize the

transdermal drug delivery by enhancing the skin penetration, it is still to be completely exploited. Hence immense investigation therefore required to be performed into the modification of HA so as to ensure more effective targeted disruption of lipid layers while protecting deeper skin layers. Further investigation on HA will thus led in the development of novel patches that will be cheaper, painless and more effective in skin penetration.

## REFERENCES

- Pegoraro C, MacNeil S, Battaglia G. Transdermal drug delivery: from micro to nano. *Nanoscale*. 2012; 4:1881-1894.
- Tahara Y, Honda S, Kamiya N, Piao H, Hirata A, Hayakawa E, Fujii T, Goto M. A solid-in-oil nanodispersion for transcutaneous protein delivery. *J. Controlled Release*. 2008; 131:14-18.
- Rein H, Experimental electroosmotic studies on living human cell, *Zeitschrift fur Biologie*, 1924; 81:125-140.
- Michaels AS, Chandrasekaran SK, Shaw JE, Drug permeation through human skin: Theory and in vitro experimental measurement, *AIChE Journal*, 1975; 21:985-996.
- Lapcik L, Lapčik L, De Smedt S, Demeester J, Chabreček, P, Hyaluronan: Preparation, Structure, Properties, and Applications, *Chemical Reviews*, 1998; 98:2663-2684.
- Dosio F, Arpicco S, Stella B, Fattal E, Hyaluronic acid for anticancer drug and nucleic acid delivery, *Advanced Drug Delivery Reviews*, 2016; 97:204-236.
- Witting, M., Boreham, A., Brodewolf, R., Vavrova, K., Alexiev, U., Friess, W., Hedtrich, S., Interactions of Hyaluronic Acid with the Skin and Implications for the Dermal Delivery of Biomacromolecules, *Molecular Pharmaceutics*, 2015; 12:1391-1401.
- Yang JA, Kim ES, Kwon JH, Kim H, Shin JH, Yun SH et al., Transdermal delivery of hyaluronic acid - Human growth hormone conjugate, *Biomaterials*, 2012; 33:5947-5954.
- El-Refaie WM, Elnaggar YSR, El-Massik MA, Abdallah OY, Novel Self-assembled, Gel-core Hyalurosomes for Non-invasive Management of Osteoarthritis: In-vitro optimization, Ex-vivo and In-vivo Permeation, *Pharmaceutical Research*, 2015; 32:2901-2911.
- Lim HJ, Cho EC, Lee JA, Kim J, A novel approach for the use of hyaluronic acid-based hydrogel nanoparticles as effective carriers for transdermal delivery systems, *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 2012; 402:80-87.
- Wei S, Xie J, Luo Y, Ma Y, Tang S, Yue P, Yang M, Hyaluronic acid based nanocrystals hydrogels for enhanced topical delivery of drug: A case study, *Carbohydrate Polymers*, 2018; 202:64-71.
- Cilurzo F, Vistoli G, Gennari CGM, Selmin F, Gardoni F, Franzè S et al., The Role of the Conformational Profile of Polysaccharides on Skin Penetration: The Case of Hyaluronan and Its Sulfates, *Chemistry & Biodiversity*, 2014; 11:551-561.
- Franzè S, Marengo A, Stella B, Minghetti P, Arpicco S, Cilurzo F, Hyaluronan-decorated liposomes as drug delivery systems for cutaneous administration, *International Journal of Pharmaceutics*, 2018; 535:333-339.
- Gao Y, Cheng X, Wang Z, Wang J, Gao T, Li P, Transdermal delivery of 10,11-Methylenedioxycompotothecin by hyaluronic acid based nanoemulsion for inhibition of keloid fibroblast, *Carbohydrate Polymers*, 2014; 112:376-386.
- Kong M, Park HJ, Stability investigation of hyaluronic acid based nanoemulsion and its potential as transdermal carrier, *Carbohydrate Polymers*, 2011; 83:1303-1310.
- Kong M., Chen XG, Kweon DK, Park HJ, Investigations on skin permeation of hyaluronic acid based nanoemulsion as transdermal carrier, *Carbohydrate Polymers*, 2011; 86: 837-843.
- Jung HS, Kong WH, Sung DK, Lee MY, Beack SE, Keum DH et al., Nanographene Oxide\_Hyaluronic Acid Conjugate for Photothermal Ablation Therapy of Skin Cancer, *ACS Nano*, 2014; 8:260-268.
- Kim KS, Kim H, Park Y, Kong WH, Lee SW, Kwok SJJ et al., Noninvasive Transdermal Vaccination Using Hyaluronan Nanocarriers and Laser Adjuvant, *Advanced Functional Materials*, 2016; 26:2512-2522.
- Kong M, Hou L, Wang J, Feng C, Liu Y, Cheng X et al., Enhanced transdermal lymphatic drug delivery of hyaluronic acid modified transfersomes for tumor metastasis therapy, *Chemical Communications*, 2015; 51:1453-1456.
- Kong M, Park H, Feng C, Hou L, Cheng X, Chen X, Construction of hyaluronic acid noisome as functional transdermal nanocarrier for tumor therapy, *Carbohydrate Polymers*, 2013; 94:634-641.
- Kim AR, Lee SL, Park SN, Properties and in vitro drug release of pH- and temperaturesensitive double cross-linked interpenetrating polymer network hydrogels based on hyaluronic acid/poly (N-isopropylacrylamide) for transdermal delivery of luteolin, *International Journal of Biological Macromolecules*, 2018; 118:731-740.
- Manca ML, Castangia I, Zaru M, Nacher A, Valenti D, Fernández-Busquets X et al., Development of curcumin loaded sodium hyaluronate immobilized vesicles (hyalurosomes) and their potential on skin inflammation and wound restoring, *Biomaterials*, 2015; 71:100-109.
- Martin M, Azoia NG, Shimanovich U, Matamá T, Gomes AC, Silva C et al., Design of Novel BSA/Hyaluronic Acid Nanodispersions for Transdermal Pharma Purposes, *Molecular Pharmaceutics*, 2014; 11:1479-1488.
- Šmejkalová D, Muthný T, Nešporová K, Hermannová M, Achbergerová E, Huerta-Angeles G et al., Hyaluronan polymeric micelles for topical drug delivery, *Carbohydrate Polymers*, 2017; 156:86-96.
- Berkó S, Maroda M, Bodnár M, Erős G, Hartmann P, Szentner K et al., Advantages of cross-linked versus linear hyaluronic acid for semisolid skin delivery systems, *European Polymer Journal*, 2013; 49:2511-2517.
- Kong BJ, Kim A, Park SN, Properties and in vitro drug release of hyaluronic acid-hydroxyethyl cellulose hydrogels for transdermal delivery of isoliquiritigenin, *Carbohydrate Polymers*, 2016; 147:473-481.
- Xie J, Ji Y, Xue W, Ma D, Hu Y, Hyaluronic acid-containing ethosomes as a potential carrier for transdermal drug delivery, *Colloids and Surfaces B: Biointerfaces*, 2018; 172:323-329.
- Yang JA, Kim ES, Kwon JH, Kim H, Shin JH, Yun SH et al., Transdermal delivery of hyaluronic acid e Human growth hormone conjugate, *Biomaterials*, 2012; 33:5947-5954.
- Yue Y, Zhao D, Yin Q, Hyaluronic acid modified nanostructured lipid carriers for transdermal bupivacaine delivery: In vitro and in vivo anesthesia evaluation, *Biomedicine & Pharmacotherapy*, 2018; 98:813-820.
- Choi JT, Park SJ, Park JH, Microneedles containing cross-linked hyaluronic acid particulates for control of degradation and swelling behavior after administration into skin, *Journal of Drug Targeting*, 2018; 26:884-894.
- Kim S, Lee J, Shayan FL, Kim S, Huh I, Ma Y et al., Physicochemical study of ascorbic acid 2-glucoside loaded hyaluronic acid dissolving microneedles irradiated by electron beam and gamma ray, *Carbohydrate Polymers*, 2018; 180:297-303.
- Matsuo K, Okamoto H, Kawai Y, Quan Y, Kamiyama F, Hirobe S et al., Vaccine efficacy of transcutaneous immunization with amyloid  $\beta$  using a dissolving microneedle array in a mouse model of Alzheimer's disease, 2014; 266:1-11
- Wu D, Quan Y, Kamiyama F, Kusamori K, Katsumi H, Sakane T, et al., Improvement of Transdermal Delivery of Sumatriptan Succinate Using a Novel Self-dissolving Microneedle Array Fabricated from Sodium Hyaluronate in Rats, *Biological and Pharmaceutical Bulletin*, 2015; 38:365-373.
- Liu S, Wu D, Quan YS, Kamiyama F, Kusamori K, Katsumi H et al., Improvement of Transdermal Delivery of Exendin -4 Using Novel Tip-Loaded Microneedle Arrays Fabricated from Hyaluronic Acid, *Molecular Pharmaceutics*, 2016; 13:272-279.
- Liu S, Jin MN, Quan YS, Kamiyama F, Katsumi H, Sakane T et al., The development and characteristics of novel microneedle arrays fabricated from hyaluronic acid, and their application in the transdermal delivery of insulin, *Journal of Controlled Release*, 2012; 161:933-941.
- Yu W, Jiang G, Zhang Y, Liu D, Xu B, Zhou J, Polymer microneedles fabricated from alginate and hyaluronate for transdermal delivery of insulin *Materials Science and Engineering: C*, 2017; 80:187-196.