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THE FUTURE OF ERECTILE DYSFUNCTION (ED)

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Summary.- *About 30-40 % of ED patients are non-responders to PDE 5 inhibitor monotherapy. Lifestyle modifications and physical activity with weight loss enhance PDE 5 inhibitor responsiveness. The same applies for combination therapies such PDE 5 inhibitors + L-Arginine 3.000mg, PDE 5 inhibitors + statins and PDE 5 inhibitors + Yohimbine. Combination of daily dosing with Tadalafil 5 mg and on demand application of sildenafil or vardenafil can improve responsiveness and erection hardness (personal experiences).*

Guanylate cyclase activators or RhoA-kinase inhibitors, either as monotherapy or in combination with PDE 5 inhibitors have shown in preclinical settings the potential to improve erectile function and represent targets for new ED drugs in the future. Immunophilin ligands were able to ameliorate erectile function after cavernous nerve injury due to pelvic surgery.

Although having shown convincing efficacy both in animals and humans the centrally acting Melanocortin Receptor (MCR) Agonists were given up for ED treatment because of unfavorable side-effects.

Promising targets for ED therapy in the future is gene therapy with several targets as well as stem cell therapy with adipose-derived or muscle-derived stem cells.

Keywords: *Erectile dysfunction. PDE 5 inhibitors. Non-responders, Guanylate cyclase activators. RhoA. kinase inhibitors. Melanocortin agonists. Gene therapy. Stem cell therapy.*

Resumen.- *Cerca del 30-40% de los pacientes con disfunción eréctil (DE) no responden a inhibidores de la PDE 5 en monoterapia. Las modificaciones del estilo de vida y la actividad física con pérdida de peso mejora la respuesta a inhibidores de la PDE 5. Lo mismo sirve para los tratamientos combinados tales como Inhibidores de la PDE 5 + L arginina 3 g, Inhibidores de la PDE 5 + estatinas e inhibidores de la PDE 5 + yohimbina. La combinación de una dosis diaria de Tadalafilo 5 mg y la aplicación de sildenafilo o vardenafilo a demanda puede mejorar la respuesta y la rigidez de las erecciones (experiencia personal).*

Los activadores de la guanilato ciclasa o los inhibidores de la Rho A kinasa, tanto en monoterapia como en combinación con los inhibidores de la PDE 5 han demostrado en escenarios preclínicos el potencial de mejorar la función eréctil y representan dianas para los nuevos fármacos en disfunción eréctil en el futuro.

Los ligandos de inmunofilina fueron capaces de mejorar la función eréctil después de la lesión de los nervios cavernoso debida a cirugía pélvica.

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Aunque han mostrado una eficacia convincente tanto en animales como en humanos los agonistas del receptor de melanocortina de acción central fueron abandonados en el tratamiento de la DE debido a efectos secundarios desfavorables.

En el futuro son dianas prometedoras en el tratamiento de la DE la terapia génica, con varias dianas, y también el tratamiento con células madre derivadas de tejido graso o muscular.

Palabras clave: *Disfunción eréctil. Inhibidores de la PDE 5. No respondedores. Activadores de la guanilato ciclasa. Inhibidores de Rho A kinasa. Agonistas de melanocortina. Terapia génica. Terapia con c*

INTRODUCTION

The successful introduction of effective oral drug therapy in the management of erectile dysfunction 1998 with Sildenafil (Viagra®) being the first representative of PDE 5 inhibitors has revolutionized the whole field of sexual dysfunction. Although the advent of this new compound class has made treatment of ED supposedly easy for both the physicians, prescribing these drugs and the patients taking them in joyful expectations, some problems remain to be solved:

1st: PDE 5 inhibitors are not effective in everybody. Considering the meanwhile huge database on all 3 older PDE 5 inhibitors sildenafil, tadalafil and vardenafil it turned out that their efficacy is limited and only between 60 and 70 % of patients are able to get erections firm enough to engage in and complete sexual intercourse. That means that for about 30-40 % of all ED patients PDE 5 inhibitors are the wrong choice.

2nd: There are clear contraindications for the use of PDE 5 inhibitors, especially in patients on medications, containing either nitrates, molsidomine or other drugs with NO donor properties.

3rd: Despite a proven broad-spectrum efficacy in the daily practice setting long-term use of PDE 5 inhibitors has resulted in similar drop-out rates of between 30 and 50 % as has been shown with previous intracavernous injection therapy (1,2). Main reasons for discontinuing PDE 5 inhibitor use were high costs, efficacy below expectancy, loss of interest in sex, need for scheduling sexual activities and partner issues (1).

All these shortcomings associated with the use of PDE 5 inhibitors indicate an urgent need for further improvement in the management of ED and motivate both independent researchers and the pharmaceutical industry to find new treatment modalities for the future, which may be even more effective or more convenient than the current use of PDE 5 inhibitors. Therefore the following chapters are dealing with both optimizing the management of ED with already available ED therapies/strategies and future targets/compounds which have already shown or may have a clear therapeutic potential in the management of ED.

Lifestyle Modifications in the Management of ED

At the latest since the publication of Esposito et al who were able to improve significantly erectile function (IIEF-EF increase of > 3) in men with ED by only achieving a loss of 10% or more in their total body weight by reducing caloric intake and increasing their level of physical activity it became evident that just lifestyle modifications are able to restore erectile function in overweight patients (3). In another recently published trial a well-defined low calorie-diet with a commercially available meal replacement (Kicstart) weight loss of ~10% in obese men with body mass index ≥ 30 kg m^{-2} and waist circumference (WC) ≥ 102 cm was significantly associated with increased insulin sensitivity, plasma testosterone levels, IIEF-5 and SDI scores, as well as reduced WC and IPSS scores, in diabetic as well as non-diabetic men. The degree of weight loss was significantly associated with improvements in plasma testosterone levels (r=-0.34), erectile function (r=-0.26) and LUTS (r=0.65) (4). In a recently published randomized controlled study the combination of physical activity (3,4 h/week) and PDE5 inhibitor treatment was statistically significant superior to PDE5 inhibitor therapy alone with IIEF restoration of ED in 77,8 % vs. 39,3 % (5).

New PDE 5 Inhibitors

Since the launch of the "old" PDE 5 Inhibitors 1998 and 2003 some other newer PDE 5 inhibitors were developed and are meanwhile available in some countries world-wide such as South Korea ,Brazil and other. These PDE 5 inhibitors are Lodenafil, Udenafil, and Mirodenafil.

Regarding the pharmacokinetic profiles Sildenafil, Vardenafil, Lodenafil and Mirodenafil are short-acting PDE 5 inhibitors with half-life times of between 2,4 and 3,9 hours whereas Mirodenafil

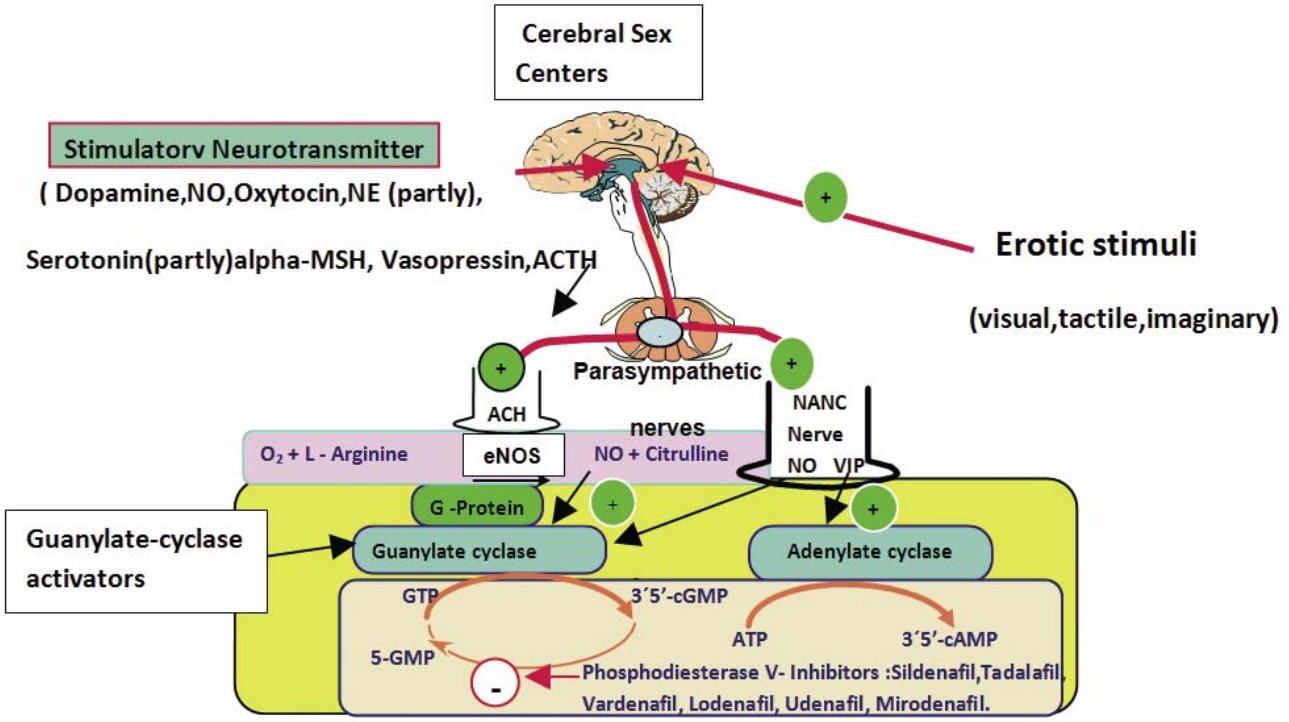

FIGURE 1.

and Tadalafil show considerable longer half-live times which translates in clearly longer duration of clinical efficacy. In this regard Tadalafil has shown in clinical trials even after 36 hours SEP-3 rates of about 60 % as compared to 54 % after 12 hours with Mirodenafil (table 1).In terms of the clinical efficacy assessed by the SEP 3 rates all 6 PDE 5 inhibitors have shown very similar and thus comparable data (Table I).

PDE 5 Inhibitors and Oral Combination Therapy

As mentioned before in more than 30 % of ED patients PDE 5 inhibitors given as mono-therapy turned out to ineffective. Some recent studies have shown that L-Arginine administered alone in doses of up to 3.000 mg/day was able to improve significantly erectile function with an increase of the IIEF-EF score

TABLE I. PHARMACOKINETICS AND SEP 3 RATES (MAINTENANCE OF ERECTION WITH COMPLETION OF SEXUAL INTERCOURSE) OF CURRENTLY APPROVED PDE 5 INHIBITORS. (LITERATURE: FOR SILDENAFIL,TADALAFIL AND VARDENAFIL: 6-9 , LODENAFIL: 10 , MIRODENAFIL:11, UDENAFIL: 12-13).

Drug and Dose	T Max (min.)	T 1/2 (h)	No.Pts.	SEP 3 (%)
Sildenafil 100 mg	70 (30-120)	3,82 ± 0,84	1720	66
Tadalafil 20 mg	120 (30-720)	17,5	1112	75
Vardenafil 20 mg	40 (15-180)	3,94 ± 1,31	601 and 864	75 and 67
Lodenafil 80 mg	72	2,4	350	66
Mirodenafil 100 mg	85	2,5	223	67
Udenafil 200 mg	60 - 90	11 - 13	167	75
Udenafil 100 mg			104	54 after 12 h

from 15,2 to 27,1 as compared to from 15,1 to 19,0 with placebo after 6 months continuous therapy (14) or has increased the efficacy when added to vardenafil 20 mg in insulin dependent diabetic men with ED (15).Our personal experiences support these findings by combining daily dosing of 3000 mg L-Arginine either with on demand PDE 5 inhibitors or daily dosing of 5 mg tadalafil in those patients who were dissatisfied with the outcome of either PDE 5 inhibitor mono-therapy. Similar observations apply for patients with the combination therapy of PDE 5 inhibitors and yohimbine, which has recently shown to relax the human corpus cavernosum through a non-adrenergic mechanism involving the activation of K+ATP-dependent channels (16). Adding statins to PDE 5 inhibitor therapy in men suffering from ED and dyslipidemia can increase the positive effect on erectile function (17,18)

Finally the personal experiences have shown that in some patients in whom neither on demand therapy with the highest dose of either PDE 5 inhibitor nor daily dosing regimen of Tadalafil 5 mg is able to induce satisfactory erections, the combination of both, i.e daily dosing of 5 mg Tadalafil and on-demand application of a short acting PDE 5 inhibitor -here sildenafil or vardenafil-, is able to convert non-responders to either therapy to responders to combination therapy.

OTHER NEW PERIPHERALLY ACTING DRUGS

Guanylate Cyclase (GC) Activators

Guanylate (syn.:guanylyl)cyclase (GC) ,here GC-B, a membrane bound enzyme, is involved in the physiological process of erection by promoting the cleavage of cGMP from GTP which subsequently triggers the erection cascade (Figure 1). Meanwhile several compounds have been developed in the laboratories of various pharmaceutical companies that activate sGC independent of NO release. Recent studies have emphasized the potential use of heme-dependent **sGC stimulators** (e.g. YC-1, BAY 41-2272, BAY 41-8543, BAY 63-2521, CFM-1571 and A-350619) and heme-independent **sGC activators** (e.g. BAY 58-2667, HMR-1766, S-3448, A-778935) in the treatment of cardiovascular diseases including erectile dysfunction (19). Because these guanylate cyclase activators are able to increase the intracellular cGMP concentration in an NO-independent mechanism it is conceivable that this class of compounds may show a higher efficacy profile than the currently used PDE 5 inhibitors which need for their efficacy profile the NO involvement. Currently a new dually acting compound has been developed by Bayer Schering Pharma, Germany, which combines the effects of a PDE 5 inhibitor (here vardenafil) with those of a GC activator (20) and is currently subject to a clinical investigational program.

RhoA-kinase inhibitors

A variety of research findings, emphasizes the role of Rho-kinase in the regulation of the corpus cavernosum smooth muscle tone, i.e. maintaining contraction and thereby preventing erection (22) It has been shown that Rho-kinase expression and activity is clearly up-regulated both in diabetes and hypertension and in hypoxemic conditions (23-25). Elevated RhoA/Rho-Kinase activity has recently been shown in the hyperlipidemic rabbit penis and is suspected to contribute to hyperlipidemia-associated ED (26).

In the animal model Rho-kinase inhibitors were able to ameliorate ED (23,24,27). Recent studies performed in hypertensive rats have provided evidence that the combination of PDE 5 inhibitors and Rho kinase inhibitors are superior to the respective mono-therapies. Finally the topical application of the Rho-kinase inhibitor Y-27632 to the rat tunica albuginea resulted in an erectile response (24). Although in the preclinical research various Rho-Kinase inhibitors have shown a promising efficacy, no one Rho-Kinase inhibitor has been investigated so far in human trials in the indication of ED.

Other peripherally acting drugs

There are many peripherally acting compounds which have shown both in animal models and in in-vitro trials the potential to improve erectile function. Among those compounds are endothelin antagonists and angiotensin II antagonists (28,29), but none of these compounds was investigated in a phase I-III clinical development program.

Compounds for cavernous nerve regeneration

Cavernous nerve injury is the predominantly etiological factor contributing to the manifestation of ED after major pelvic surgery (radical prostatectomy, cystectomy, rectum amputation): **Immunophilin ligands** provide potentially new alternatives for the treatment of post pelvic surgery ED. A review of available reports of studies investigating the effects and neurotrophic mechanisms of immunophilin ligands has shown that treatment with prototype immunosuppressive immunophilin ligands FK506 (FK)

and rapamycin (Rapa) was able to improve erectile function in animal models of cavernous nerve injury. Similarly, non-immunosuppressive analogs such as GPI-1046 and FK1706 were effective in recovery of erections after cavernous nerve injury. Neuronal nitric oxide may influence the erection recovery effects of immunophilin ligands after CN injury and antioxidative actions of immunophilin ligands contribute to their neurotrophic effects (30). Although successful in the animal setting a huge human placebo-controlled randomized study, involving 131 men (mean age 55 ± 6 years) undergoing bilateral nerve sparing RRP, was unable to show significant effects of the immunosuppressant immunophilin tacrolimus, on recovery of erectile function after 3 months as compared to placebo (31). Another compound, **sonic hedgehog (SHH)** protein, delivered by noodle nanotechnology, has been effective in speeding cavernous nerve regeneration in the rat cavernous neurotomy model (32).

CENTRALLY ACTING DRUGS

Melanocortin Receptor (MCR) Agonists

Presently there are 5 MCRs identified, which are all 5 activated by adreno-corticotropin (ACTH) and 4 out of 5, except MCR 2, by alpha melanocyte stimulating hormone (alpha-MSH). Of the 5 MCRs only two (MC3R and MC4R) are expressed in cerebral regions known to be involved in the modulation of erectile function. **Melanotan II**, a cyclic peptide analogue of α-MSH, has been a long time under investigation for its usefulness in the management of ED both as subcutaneous injection and intranasal application.

Although the results of phase II/III trials both with intranasal and sc injectable Melanocortin receptor agonist PT-141 (originally **Melanotan II**), have shown improved erections in 66-67% with the 10-20 mg dose the project was finally stopped because of the side-effect profile, i.e. especially because of elevated blood pressure in some patients as well as nausea and vomiting in 10-20 % (33,43).

Other Centrally Acting Drugs

Other centrally acting drugs with proven pro-erectile profiles are **Dopamine agonists, Serotonin receptor agonists** (5 $HT_{1C/2A \text{ and } C}$), **Glutamate and Hexarelin analogues**, but none of these compounds was investigated in a clinical phase II/III program except for apomorphine, which finally was withdrawn from the market because of its inferior efficacy as compared to sildenafil (28,29).

GENE THERAPY AND TISSUE ENGINEERING IN ED

Gene Therapy

The rationale of gene therapy is to introduce new or repair/replace damaged genetic materials (DNA or RNA) into the cells of a target (i.e. the non or poorly functioning cavernous bodies) in order to recover the organ's function and restore erectile function. Without any doubt gene therapy is an attractive therapeutic option for the treatment of ED. The penis is also a convenient tissue target for gene therapy because of its external location, the ubiquity of endothelial lined spaces, and low level of blood flow.

A variety of gene therapy trials have been conducted for the treatment of ED in the past 12 years. Due to its outstanding importance in the physiology of normal erectile function, many gene therapy studies have focused on the NO/GC/cGMP pathway. Other targets for genetic manipulation in the management of ED were: nerve and vascular growth factors, (e.g. brain-derived nerve growth factor [BDNF], vascular endothelial growth factor [VEGF]), the cyclic adenosine monophosphate (cAMP) cascade (i.e., calcitonin gene-related peptide [CGRP] receptor), the calcium sensitization pathway, and K^+ channel gene expression which serves as a cellular convergence point for mediating the effects of all of the above (35,36).

At present only one gene transfer phase 1 safety trial for ED has been successfully conducted and published, using gene transfer of potassium (hMaxi-K) channels to treat smooth muscle diseases (38,39).For cost reasons this project as well as other gene therapy approaches, successfully tried in animal models, have not proceeded so far. It may be argued that gene therapy in ED, although very appealing and promising, will not make further progress in the near future, because of the cost issues linked to these projects.

Stem cell Therapy for Erectile Dysfunction

Reconstruction of normal erectile tissue, here the cavernous tissue, using autologous cells, harvested from the patient's own body, is an intriguing concept for the treatment of severe ED forms, non-responding to conventional medical therapy such as PDE5 inhibitors. Initial experiments were performed in order to determine the feasibility of creating corporal smooth muscle and endothelial cells *in vivo* using cultured human corporal smooth muscle cells seeded onto biodegradable polymers. Their results were confirmed with human corporal smooth muscle

and endothelial cells seeded on acellular matrices processed from donor rabbit corpora cavernosa (35).

Adipose-derived stem cells (ADSCs) represent a somatic stem cell population located in fat tissue that has the ability for self-renewal, differentiation into one or more phenotypes, and functional regeneration of damaged tissue. By using a stem cell-based therapy ADSCs may benefit the recovery of erectile function. A review of the currently available data has indicated that application of ADSCs, as well as other kinds

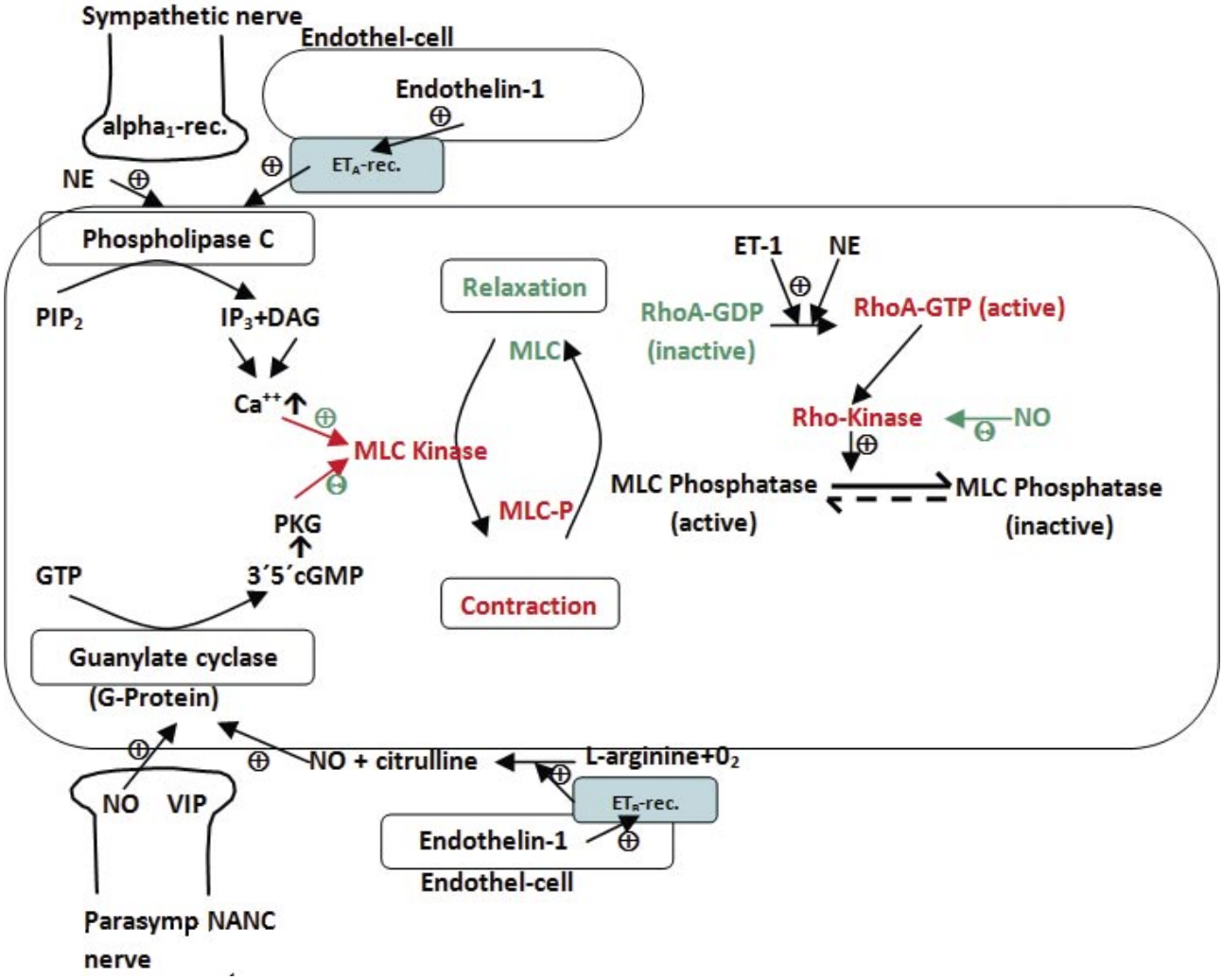

FIGURE 2. The role of Rho A and Rho-kinase in the erectile process (modified, Source Mills 21): cGMP is the most important second neurotransmitter of erection and stimulates the proteinkinase G which in turn initiates phosphorylation of membrane bound proteins at the potassium channels. This leads to potassium ion outflow into the extracellular space resulting in hyperpolarization. Hyperpolarization leads to the closure of the L-type calcium channels in turn resulting in a decrease of intracellular Ca++-ion concentrations. Physiologically intracellular Ca++ along with calmodulin activates the myosin light chain (MLC) kinase which catalyzes the phosphorylation of the myosin light chains and induces the actin-myosin interaction finally resulting in contraction of the cavernous smooth musculature and thus preventing erection. The phosphorylated myosin light chain is dephosphorylated by the active (dephosphorylated) form of MLC phosphatase with the result of corpus cavernosum relaxation and erection. Phosphorylation which means inactivation of MLC phosphatase is catalyzed by Rho-kinase. Rho A is a small GTPase which is inactive, while GDP is bound, and is active while GTP is bound. The activated RhoA stimulates the serine/threonine kinase named Rho-kinase. Rho-kinase itself phosphorylates the MLC phosphatase at the myosin binding unit and thus inactivates this enzyme resulting in smooth muscle contraction and flaccidity of the penis. The so-called Rho-kinase inhibitors, developed recently to assess their potential for the treatment of erectile dysfunction, activate the MLC phosphatase and lead to relaxation and erection.

of stem cells have under specific induction medium conditions the potential to differentiate into neuron-like cells, smooth muscle cells, and endothelium in vitro (39). In a preliminary in vivo experiment, the ADSCs functionally recovered the impaired erectile function but the exactly underlying mechanism needs to be further on examined. These preliminary data indicate that ADSCs are a potential source for stem cell-based therapies, which imply the possibility of an effective clinical therapy for ED in the near future. The same observation applies for muscle-derived stem-cells (MDSCs) which have been implanted into the corpora cavernosa of aged rats and finally corrected ED thru converting into SMCs (40).

CONCLUSION

As briefly summarized above many attractive targets have been figured out for future treatment of ED including gene therapy and tissue engineering, but both tremendous costs and safety concerns, may prevent many of these promising options from being developed for market approval. Therefore at present and in the near future, we the therapists and the patients, have to rely on what is available now and should more seriously think about lifestyle modifications and combination therapies, than dreaming about options that may never reach the market.

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