SUMMARY

Alternative therapies are proposed to be developed via two strategies: i. development of early detection methods and smart renal regeneration therapies, in order to intervene in and cure kidney diseases at an early stage, and ii. design and development of alternative renal replacement therapies, such as a bio- artificial kidney(devices) using tissue engineering strategies.

The possibility of restoration of kidney tissue using cells, regenerative factors, biomaterials, or combinations of these three, is approaching. Cell-based and factor-based approaches might be able to intervene in impaired kidney functions by induction of renal regeneration. Cell-based approach might also be applied to grow neo kidneys.Material-based strategies might be used to design and develop a bioartificial kidney (device).

Cell-based approaches: stem/progenitor cell injection:

A large number of studies have reported on the contribution of BMDC to renal regeneration. These cells might be injected in the kidney or in the blood to ultimately end up in the impaired kidney.

Several studies have shown that BMDC after damage can replace tubular cells, mesangial cells , podocytes , interstitial cells and endothelial cells. But except for interstitial engraftment, the relative contribution is very low and does not exceed a few percent of the total proliferating cell fraction.

There is large body of evidence for a paracrine role of BMDC in the repair process. BMDC are recruited after damage, and their incorporation rate is enhanced. Moreover, they produce a variety of paracrine factors, such as VEGF, IGF, bFGF, HGF, and TGFβ that can enhance repair ..It has been shown that almost 30% of myofibroblasts in fibrotic kidneys are bone marrow-derived.

Tight regulation of BMDC differentiation in the interstitium is therefore important. It seems that BMDC are able to give rise to multiple cell types depending on the local composition of the environment, i.e. the ECM and growth factors present. For this reason, one should be cautious using BMDC for local regeneration therapy.

An alternative strategy would be to use BMDC that have been differentiated ex vivo towards a specific renal lineage prior to implantation. Furthermore, it is important to notice that the uremic environment of ESRD patients functionally impairs progenitor cells such as endothelial progenitor cells.

Cell-based approaches: neo-kidneys:

Another interesting approach is the formation of neo-kidneys from implanted embryonic tissue, i.e. renal primordia. A great advantage of this approach is that during maturation the tissue is vascularized by the host, which improves compatibility.

However, a major obstacle is the source of embryonic tissue, which is closely connected to ethical issues. In the future, these cells might be derived from embryonic stem cells (ESC) that are induced towards the renal epithelial lineage.

In teratomas from undifferentiated murine ESC, mesonephric and ureteric bud-like structures have been observed, suggesting that differentiation towards renal lineage is possible .

A number of studies have explored the possibility to generate renal epithelia from ESC. After injection, murine ESC has been shown to integrate into the tubuli of developing kidneys. Differentiation towards a specific lineage is very complicated and depends on factors secreted in the environment of the developing kidney, this is difficult to mimic in vitro.

Nevertheless, studies have shown that ESC, after formation of embryoid bodies, were able to differentiate towards the renal epithelial lineage by administration of either BMP4, or by a combination of activin A, BMP7 and retinoic acid, or by gene transfection with the Wnt4 gene.

Also, the materials and ECM components on which ESC grow in vitro are important factors in renal tubule formation. Although this approach seems very attractive for renal regeneration and/or replacement, the use of embryonic tissues is closely connected to ethical and safety issues.

However, an exciting study showed that both renal glomerular and tubular structures could originate from single primary renal cells cultured in a three-dimensional environment .Furthermore, other studies have shown that neo-kidneys could be produced using decellularized kidneys as scaffold .It is clear from these studies that immense progress is made using cell based approaches. However the step from in-vitro to real in-vivo kidney regeneration using different renal cells or stem/progenitor cells has a long way to go.

Factor-based approaches: delivery of kidney regenerating factors:

Exogenous growth factor administration to enhance renal regeneration has been studied in large detail. A number of studies, in which up-regulation of growth factor genes in relation to renal repair processes has been found, have provided the rationale for growth factor delivery strategies.

Systemic injection of EGF and HGF successfully enhanced recovery and survival after acute kidney injury in animal models. In addition, systemic administration of IGF1 was evaluated in human trials, and has shown to improve recovery after acute kidney injury.

Furthermore, many examples have been shown in which BMP7 was delivered intravenously or intraperitoneally, and resulted in recovery of renal function in animal models.

However, a major disadvantage is that these therapies might lead to unwanted side effects in other organs especially when the drug is administered systemically.

These growth factors show pleiotropic effects, which implies that they need to be targeted to a specific site in the body. Besides that, the stability of these proteins is generally very low; therefore growth factors need to be administered repeatedly.

For example, BMP7 has a serum half-life of 30 min, and can be found in the kidney shortly after intravenous administration.

Good control over the exact growth factor concentration is therefore important. For example, a low dose of BMP7 has shown to stimulate epithelial cell proliferation, whereas a high dose has shown to inhibit proliferation and induce apoptosis in the regenerating epithelial tubule. The recovery of renal function by systemic administration of one growth factor is not very likely to reverse or alter the progression of renal disease. Correct regeneration requires the delivery of multiple growth factors in a spatiotemporally controlled way.

Therefore sophisticated local delivery and/or targeting systems need to be developed to successfully enhance renal repair.

Material-based approaches: bioartificial kidneys:

In the future, a possible solution for patients with ESRD, besides dialysis or transplantation, might be a tissue-engineered kidney. Ideally, a tissue-engineered kidney should be able to replace all kidney functions. In view

of the extremely complex renal architecture and the great variety of cell types, i.e. more than 15 different cells, bioengineering of a complete neo-kidney is virtually impossible.

Therefore, another approach is the bioengineering of an extracorporeal renal device using a membrane and a single renal cell type that has to form a monolayer of cells in order to ultimately replace critical endocrine and metabolic renal functions.

Such a bioartificial kidney might be applied as a renal assist device (RAD), and exert its function when placed in series with a conventional hemodialysis module. Many researchers have used renal epithelial cell lines from porcine or canine origin, i.e. respectively Lewis lung cancer-porcine kidney 1 (LLC-PK1) cells and Madin-Darby canine kidney (MDCK) cells.

Although, confluent layers of MDCK cells on polycarbonate membranes initially displayed active Na+ transport, functional properties could not be maintained after 2weeks .Moreover, the loss of function was associated with the aberrant distribution of Na+/K+ ATPase, multilayer growth, and necrosis.

Reabsorption of water, glucose and sodium could be maintained up to 10 days when LLC-PK1 cells were used. Importantly, the type of membrane material and the ECM coating, appeared to be critical for the adhesion and functional differentiation of renal epithelial cells .Despite the limited transport function, the activity of the renal epithelial cells could attenuate the consequences of septic shock by modulating plasma cytokine levels.

To this end, initial phase I and II clinical trials have indicated that RAD treatment for 72 h improved long-term survival and renal recovery

of patients with acute renal failure . The application of renal epithelial cells ex vivo is limited by the rapid loss of renal epithelial cell characteristics.

Exposure of renal epithelial cells to an atypical physicochemical environment, e.g. fluctuations of glucose levels, lactate accumulation, hypoxia or hyperoxia, an aberrant ECM, and lack of heterotypic cell interactions, contribute to dedifferentiation events. Renal epithelial cell cultures under organotypic perfusion culture conditions might help to maintain differentiated renal epithelial cell features.

The vast majority of studies in this field have been focused on reconstruction of the renal tubular system. However, a bioartificial tubular system is only feasible if it is also connected to a hemofiltration device. Therefore, the RAD is connected to a hemodialysis module.

Furthermore, development of a hemofiltration device which can be implanted in vivo is an extreme challenge; especially because almost every implantable material will elicit a foreign body response, which eventually might result in calcification and encapsulation of the hemofilter. Nevertheless, important progress was made through the development of a renal assist device, and an implantable hemofilter. It is a great challenge to improve these systems in such a way that the epithelial cells within the device encounter a regenerative, natural renal niche.