Evolving treatments of virus-associated HCC: new targets and drugs

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1. ABSTRACT

Hepatocellular carcinoma (HCC) is one of the most common malignant tumors worldwide. The major etiologies and risk factors for HCC development are well defined and some of the multiple steps involved in hepatocarcinogenesis have been elucidated in recent years. The therapeutic options fall into five main categories: (1) surgical interventions, incl. liver transplantation, (2) percutaneous interventions, incl. ethanol injection and radiofrequency thermal ablation, (3) transarterial interventions, (4) radiation therapy and (5) drugs as well as gene and immune therapies. Because of the poor survival of the majority of patients, HCC prevention as well as early diagnosis and the development of novel systemic therapies for advanced disease are of paramount importance. In this context, recent data indicate that the 'targeted therapy' with monoclonal antibodies (mabs) or small molecule tyrosine kinase inhibitors (nibs) and other drugs seem to be effective to some degree. New technologies, including gene expression profiling and proteomic analyses, should allow to further elucidate the molecular events underlying HCC development and to identify novel diagnostic markers as well as therapeutic and preventive targets.

2. INTRODUCTION

Hepatocellular carcinoma (HCC) is one of the most common malignant tumors worldwide and has been recently reviewed (1, 2). The incidence ranges from <10 cases per 100,000 population and year in North America and Western Europe to 50-150 cases in parts of Africa and Asia where HCC is responsible for a large proportion of cancer deaths (3). However, a rise in the incidence of and mortality from HCC, most likely reflecting the prevalence of hepatitis C virus (HCV) infection, has recently been observed also in most industrialized countries (4).

The major etiologies of HCC are well defined (Table 1) and include in addition to the well known factors an elevated body mass index in men (5) as well as diabetes mellitus (6). Some of the steps involved in the molecular pathogenesis of HCC have been elucidated in recent years. As for most types of cancer, hepatocarcinogenesis is a multistep process involving different genetic alterations that ultimately lead to malignant transformation of the hepatocyte. While significant progress has been made in recognizing the sequence of events in other forms of cancer, most notably in colorectal cancer and certain

Table 1. Major HCC etiologies

Chronic Hepatitis B or D, and C
Toxins (e.g., alcohol, tobacco, aflatoxins)
Hereditary metabolic liver diseases (e.g., hereditary hemochromatosis, α-1-antitrypsin deficiency)
Autoimmune hepatitis
States of insulin resistance
• Overweight in males
• Diabetes mellitus
• Non-alcoholic steatohepatitis (NASH) or non-alcoholic
• fatty liver disease (NAFLD)

hematopoietic malignancies, the molecular events and their interactions in hepatocarcinogenesis are still poorly understood. HCCs are phenotypically (morphology, microscopy) and genetically very heterogenous tumors, possibly reflecting in part the heterogeneity of etiologic factors implicated in HCC development, the complexity of hepatocyte functions and the late stage at which HCCs usually become clinically symptomatic and detectable. Malignant transformation of hepatocytes may occur regardless of the etiology through a pathway of increased liver cell turnover, induced by chronic liver injury and regeneration in a context of inflammation, immune response and oxidative DNA damage. This may result in genetic alterations, such as the activation of cellular oncogenes, the inactivation of tumor suppressor genes, possibly in cooperation with genomic instability, including DNA mismatch repair defects and impaired chromosomal segregation, overexpression of growth and angiogenic factors, and telomerase activation (7-11). Further, there is evidence for the existence of liver stem cells that may be progenitors of HCC cells (12). Chronic viral hepatitis B or D, C, alcohol, metabolic liver diseases such as hemochromatosis and α-1-antitrypsin deficiency as well as non-alcoholic fatty liver disease may act predominantly through this pathway of chronic liver injury, regeneration, and cirrhosis. The major clinical risk factor for HCC development is liver cirrhosis since 70-90% of HCCs develop in a cirrhotic liver. Most HCCs occur after many years or decades of chronic hepatitis that provides the mitogenic and mutagenic environment to precipitate random genetic alterations resulting in the malignant transformation of hepatocytes and HCC development.

The HCC risk in patients with liver cirrhosis depends on the activity, duration and the etiology of the underlying liver disease. Clinical and biological variables (age, anti-HCV positivity, PTT and platelet count) allow to further identify a subset of cirrhotic patients with a very high risk for HCC development (13). Coexistence of etiologies, e. g., hepatitis B virus (HBV) and HCV infection, HBV infection and aflatoxin B1 (11, 14), HBV/HCV infection and alcohol or diabetes mellitus (15), HCV infection and liver steatosis (16) or environmental factors, e.g., alcohol (11, 17, 18) as well as diabetes mellitus, obesity and tobacco (5, 18-20) increase the relative risk of HCC development of a single etiology. Also, occult HBV infection (anti-HBc positive only) carries a significant HCC risk (21, 22). Interestingly, coffee consumption appears to reduce the HCC incidence (23-25).

In general, HCCs are more frequent in males than in females and the incidence increases with age. On the other hand, there is evidence that HBV -and possibly also

HCV- may under certain circumstances play an additional direct role in the molecular pathogenesis of HCC. Finally, aflatoxins have been shown to induce mutations of the p53 tumor suppressor gene, thus pointing to the contribution of an environmental factor to tumor development at the molecular level. Further, in a transgenic mouse model it has been shown that chronic immune-mediated liver cell injury without environmental or infectious agents is sufficient to cause HCC (26) and that inhibition of cytotoxic T lymphocyte-induced apoptosis and chronic inflammation by neutralization of the Fas ligand prevents HCC development in this model (27). In addition, also in a transgenic mouse model it has been demonstrated that NF-kappaB may be the link between inflammation and HCC development (28, 29). Finally, individual polymorphisms of drug metabolizing enzymes, e.g., various cytochrome P450 oxidases, N-acetyltransferases and glutathione-S-transferase, may contribute to the genetic susceptibility to HCC development(30).

3. HCC SCREENING, STAGING AND NATURAL COURSE

HCC screening is routinely done in all individuals at risk for HCC development at 6 months intervals and includes laboratory tests, such as liver function tests and alpha fetoprotein (AFP), imaging analyses, e. g., abdominal ultrasound, contrast enhanced CT or MRI and histopathology (31). Apart from AFP two additional tumor markers are now commercially available (32): des-gamma-carboxy-prothrombin (DCP) and lens culinaris agglutinin reactive fraction of AFP (AFP-L3).

For HCC staging seven systems have been proposed that assess the extent and the prognosis of the disease (31, 33): the Okuda staging system (34), the TNM classification and its modification by the 'Union International Contre Cancer (UICC)'(35), the 'Barcelona Clinic Liver Cancer (BCLC)' classification (36), the 'Cancer of the Liver Italian Program (CLIP)' score (37), the 'Japan Integrated Staging (JIS)' score (38), the Groupe d'Etude de Traitement du Carcinoma Hepatocellulaire (GRETCH) score (39) and the Chinese University Prognostic Index (CUPI). The Okuda staging system is very effective for the identification of a subgroup of patients (Okuda III) with a very poor prognosis who should be treated with best supportive care (BSC) only. The BCLC classification appears especially useful for the selection of treatment options but has not been independently validated. The CLIP score was shown to be superior to the Okuda staging system but has not been systematically assessed in patients undergoing resection or liver transplantation (LTx). While a recent study indicates that the new prognostic JIS score is superior to the CLIP score(38), a comparison of the staging systems in an American cohort revealed that the BCLC had the best independent predictive power (40).

The natural course of the disease and the median survival of patients with HCC depend on the stage of the disease at the time of diagnosis. In patients with CLIP score O or Okuda stage I the median survival is in the range of 23-69 months, while in patients with CLIP score 3-5 or
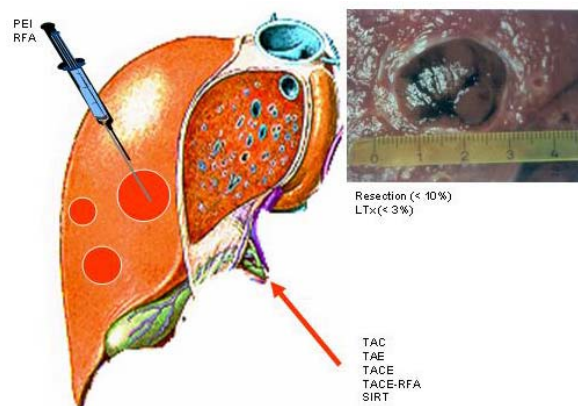
Figure 1. HCC: Current therapeutic options

Okuda stage III median survival is only 1-14 months (41). The staging system is clinically most important for the choice of the appropriate therapeutic strategy for individual patients. Cirrhotic patients developing a HCC during the last 5 years of surveillance survived longer than previously, due to improved management of the tumor and of the complications of cirrhosis (42). Importantly, however, in a population-based study in the US underutilization of potentially curative therapies even among patients with favourable HCC features is a problem that needs to be addressed (43).

4. ESTABLISHED THERAPIES

Therapies for HCC can be devided into several categories: surgical interventions (tumor resection and LTx), percutaneous interventions (ethanol injection, radiofrequency thermal ablation), transarterial interventions (embolization, chemoperfusion, chemoembolization or selective internal radiation [SIRT]), radiation therapy and drugs, including gene and immune therapy (Figure 1). Potentially curative therapies are tumor resection, LTx, and percutaneous interventions that can result in a complete response and improved survival in a high proportion of patients. In selected cases transarterial interventions results in palliation with in some cases good response rates and improved survival. Except for sorafenib, drugs as well as conventional radiotherapy have no proven efficacy.

To date, surgical, percutaneous and transarterial interventions have not been compared in randomized controlled trials. Tumor resection and LTx result in selected patient populations in 5-year survival rates of 60-70%, with LTx being the best treatment for patients with single lesions and advanced liver disease, e.g., decompensated cirrhosis, or multicentric small tumours. Percutaneous interventions, again in selected patient populations, result in 5-year survival rates of 40-50%. In the following the different therapeutic options as well as primary and secondary HCC prevention will be discussed in some detail.

4.1. Resection

In patients without concomitant liver cirrhosis (5% in Western countries, 40% in Subsahara Africa and Asia) HCC resection is the treatment of choice with low rates of life-threatening complications. By comparison, in the majority of patients with cirrhosis, strict selection is required to avoid resection-related complications, especially postoperative liver failure (44). Apart from bilirubin and albumin concentration as well as platelet count and indocyanine green clearance (45, 46), a recent study identfied an elevated serum concentration of 7s-collagen as an independent risk factor of postoperative liver failure (47).

Resection-related mortality should be <1-3%, and the 5-year survival rates should be >50%. In patients with normal liver function (normal indocyanine green retention rate and bilirubin level), absence of clinically relevant portal hypertension (hepatic venous pressure gradient <10 mm Hg, no esophageal varices, no splenomegaly, platelet counts >100 x 10^9/ L) and one asymptomatic HCC lesion only, 5-year survival rates of 70% can be achieved. By comparison in patients with clinically relevant portal hypertension, 5-year survival rates are about 50% only and in patients with portal hypertension and evidence of impaired liver function, 5-year-survival rates are even lower.

After successful HCC resection tumor recurrence in the cirrhotic liver (local recurrence as well as *de novo* tumors) in about 70% of patients at 5 years is a major clinical problem. The risk of recurrence is especially high in patients with microvascular invasion and/ or additional tumor nodules (45, 48). Therefore, strategies aimed at secondary HCC prevention are of paramount importance (see below).

4.2. Liver transplantation

LTx is in principle the optimal therapeutic option for HCCs because it simultaneously removes the tumour and the underlying cirrhosis, including the risk of HCC recurrence (44, 49-53). While broad selection criteria applied previously led to poor results with recurrence rates of about 50% and 5-year survival rates <40%, the current criteria for LTx in patients with HCC (1 lesion <5 cm in diameter or maximum 3 lesions < 3 cm in diameter) result in 5-year survival rates of 70% and more and recurrence rates <15% (54-56). Possibly these criteria can be extended in the future, depending on more experience based on the stage of the disease, macrovascular invasion, histopathological characteristics (histopathology, aneuploidy, microvascular invasion) as well as DNA and RNA chip data (molecular signature, proteomic signature and others) (57, 58).

Clinically, it is most important to shorten the waiting time for LTx to <6 months. This is difficult to achieve with cadaveric LTx given the shortage of donors. With a waiting time >12 months in some Western countries, the drop-out rate of patients is 20-50%. To bridge the time to LTx and to prevent tumor progression, neoadjuvant treatment, e. g., percutaneous and transarterial interventions may lead to an improved outcome (59) (see below). While marginal livers, domino donors, and split LTx had no major impact, living donor LTx has been

shown to be an alternative to cadaveric LTx. Around 3,000 interventions have been done worldwide. However, living donor LTx is a complex procedure that is associated with a morbidity of 20-40% and a donor mortality of 0.3-0.5% (56, 60, 61). Therefore, a very careful selection of patients and donors, including the consideration of ethical, societal and legal issues are central to the successful implementation of living donor LTx for the treatment of patients with HCCs (62).

4.3. Percutaneous interventions

Percutaneous interventions are the best options for small unresectable HCCs (63-65). Tumor ablations can be achieved chemically by percutaneous ethanol injection (PEI) or acetic acid injection (PAI) or thermally by radiofrequency thermal ablation (RFA), microwave-heat induced thermotherapy (HiTT), laser induced thermotherapy (LiTT), or cryoablation. Apart from percutaneous interventions, these techniques can be applied also laparoscopically or after laparotomy.

Initially, PEI was the most widely used percutaneous intervention (66, 67). It is safe, easy to perform, inexpensive and achieves complete tumor response rates of 90-100% in HCCs smaller than 2 cm in diameter, 70% in HCCs of 3 cm diameter and 50% in HCCs of 5 cm in diameter. Patients with liver cirrhosis Child A with complete responses can achieve 5-year survival rates of 50% and more (68). Therefore, PEI is procedure of choice for patients with a single HCC lesion smaller than 5 cm in diameter or with up to 3 lesions smaller than 3 cm in diameter. Survival is predicted by the initial response to PEI (69). However, a recent comparative study demonstrated that LTx is superior to PEI (53).

RFA is an alternative to PEI and is now the most widely used percutaneous therapy (64, 65, 70, 71). Several devices are available that can applied percutaneously, laparoscopically, or during laparotomy. The efficacy of RFA is similar to PEI but requires generally only a single session (72). While more expensive than PEI, RFA offers a better local tumor control and the potential advantage of allowing the ablation of tumors larger than 5 cm, especially with newer generation devices. However, 5-year survival rates after complete response to RFA are currently, similar to PEI, around 30-40%, depending among others on the Child stage of the underlying liver cirrhosis. In a review of 3670 patients treated by RFA, mortality was 0-5% and the complication rate 8-9% (73). A systemic review of randomised controlled trials for HCC tretated with percutaneous ablation therapies, RFA demonstrated a significant improved 3-year survival as compared to patients treated with PEI (74). Predictors of treatment response are tumour size and morphology (well encapsulated versus invasive). While safe and in case of a complete response highly effective with a prolonged survival (75), RFA is associated with a high risk of tumor persistence in the targeted nodule (76). Therefore, except for very small HCCs (< 2cm) (77) in a curative setting RFA should not be considered as an independent HCC therapy but rather as a bridging strategy for LTx, for example .

Another non-invasive thermal HCC ablation is based on MRI-guided high intensity focused ultrasound (HIFU) (78). While there is relatively little experience with this technique to date, this method may eventually prove clinically useful, possibly in combination with a transarterial intervention (see below).

Interestingly, comparing percutaneous ablation and surgery, recent evidence suggests that RFA is as effective as resection for the treatment of small HCCs (77, 79). The combination of RFA with transarterial interventions is discussed below.

Taken together, percutaneous HCC ablation by PEI and/ or RFA is an effective treatment for patients with HCCs that prolongs tumor-free and overall survival, especially if surgery is not feasible. This strategy is now being evaluated also for the treatment of liver metastases (80).

4.4. Transarterial interventions

Transarterial embolization (TAE), chemoperfusion (TAC) and chemoembolization (TACE) are the most widely used treatments for HCCs that are unresectable or cannot be effectively treated by percutaneous interventions (81-84). Embolization agents may be administered alone (embolization) or after selective intra-arterial chemotherapy (generally doxorubicin, mitomycin or cisplatin) mixed with lipiodol (chemoembolization). Recent evidence further suggests that transarterial injection of drug eluting beads is also highly effective.

TAE or TACE results in partial responses in 15-55% of patients, delays tumour progression and vascular invasion and results in a survival benefit compared with conservative management. The most important aspect is the selection of patients, i.e., patients should have preserved liver function (Child A) and asymptomatic multinodular tumours without vascular invasion or extrahepatic spread (82, 85). In a prospective study in 8,510 patients the 5-and 7-year survival was 26% and 16%, respectively, the severity of the underlying liver disease, tumor stage, and AFP level being independent prognostic factors (86). In patients with advanced liver disease (Child B or C), however, treatment-induced liver failure may offset the anti-tumor effect or survival benefit of the intervention.

In a randomized controlled clinical study the combination of TACE and PEI/ RFA improved the survival of patients with HCC Okuda stage I, as compared to TACE alone (87-89). Further in a pilot study, intraarterial doxorubicin drug-eluting beads seem to enhance the effect of RFA (90).

In an adjuvant setting, postoperative TACE may improve survival in patients with risk factors for residual tumor (91).

A novel treatment concept is transarterial ethanol ablation (TEA) based on a lipiodol-ethanol mixture that contains 33% ethanol by volume and causes endothelial
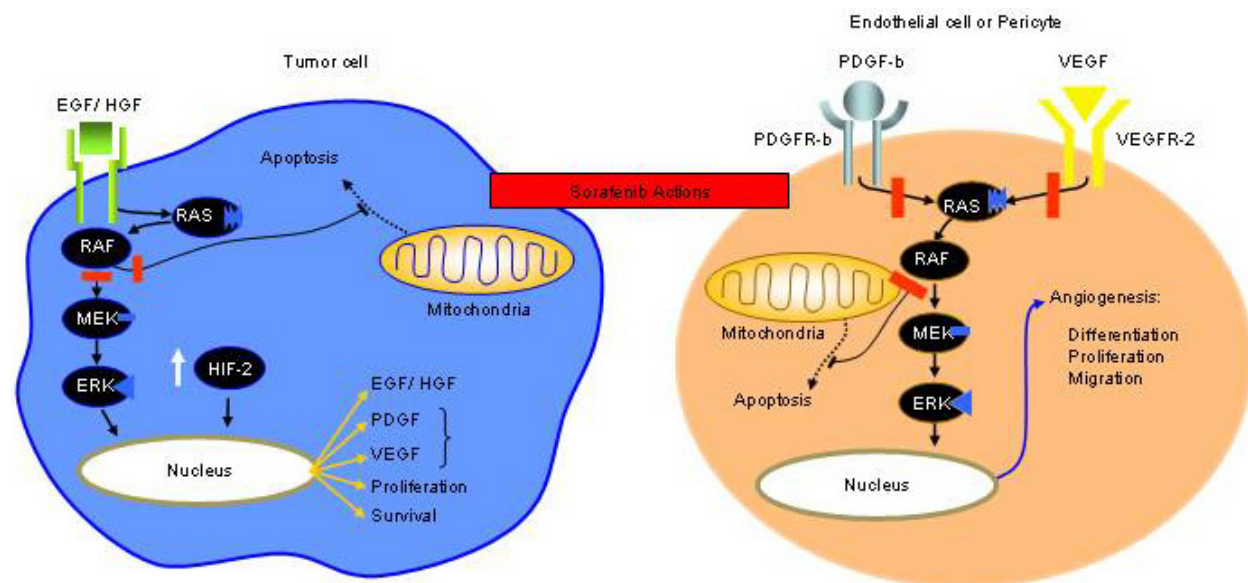
Figure 2. Action of tyrosine kinase inhibitors (according to (160))

damage and thrombosis of the arteriolar lumen of the tumor feeding vessels, resulting in tumor infarction. In a case-controlled study including 60 patients TEA was superior to TACE (92).

4.5. Drugs

To date sorafenib is the only drug that has been approved for the treatment of HCC. It is a multikinase inhibitor which effectively blocks the *raf* pathway, resulting in a block of apoptosis resistance, angiogenesis, proliferation and invasion/ metastasis (Figure 2). In a double-blind, randomized, placebo-controlled trial 800 mg sorafenib/ d or placebo was given to a total of 602 patients with liver cirrhosis Child A and advanced HCC. Sorafenib significantly prolonged overall survival (10.7 vs. 7.9 months) with acceptable toxicity (mainly diarrhea and hand-foot-syndrome) (93). While effective to some degree, it certainly can and should not be considered a routine therapy for advanced HCC, given the limited efficacy, severe side effects and high costs (94). Nevertheless, sorafenib is a benchmark drug with the perspective for further improvement of HCC treatment.

5. EVOLVING THERAPIES

5.1. Optimization of established therapies

Recently, in a pilot study a novel drug formulation based on drug eluting beads has been used for TACE and resulted in lower systemic drug levels and reduced toxicity (95).

5.2. Radiation therapy

While radiation therapy has played a minor role in the primary treatment of HCC to date, selective intra-arterial injection of [131]iodine-labeled lipiodol has been performed in some patients (96) but needs further clinical evaluation before a recommendation can be made. Further, high dose proton beam radiotherapy and external beam radiation as well as Yttrium-90 microsphere treatment, selective internal radiation therapy (SIRT), have been recently explored in clinical trials in patients with unresectable HCC (97-100), also with the intention to downstage the disease or bridge to transplantation (101). These strategies will certainly be further evaluated in clinical studies and may become a treatment option in the future.

5.3. Drugs

A number of systemic chemotherapies, hormones and other drugs (Table 2) have been evaluated in clinical trials (102-104). While most chemotherapeutic agents, tamoxifen (105), octreotide (106, 107) and interferon (81) have not been shown to be effective in randomized controlled clinical trials, there are a number of substances that may deserve further clinical evaluation, e.g., gemcitabine (108, 109), thymostimulin (110), α-1-thymosin (111), pravastatin (112), thalidomide (113), megestrol acetate (114), Cox-2 inhibitors (115) in combination with capecitabine, pamidronate (116) and others. Further, in an animal model, troglitazone resulted in an impressive reduction of HCC growth (117).

At present the most promising drugs are several small molecule tyrosine kinase inhibitors (nibs), e.g., sorafenib (see above), erlotinib, gefitinib (118) as well as monoclonal antibodies (mabs), e.g., bevacizumab or cetuximab and other drug classes, incl. the proteasome inhibitor bortezomib (Table 3) (119-122). While sorafenib has been approved for the treatment of HCC, given the limited efficacy, severe side effects and high costs, further agents alone or in combination with other novel or established strategies, e.g., RFA and/ or TACE are needed to offer a therapeutic option to the majority of patients with

Table 2. Drugs evaluated in patients with HCC

Chemotherapeutic Agents
• 5-Fluorouracil
• Capecitabine
• Gemcitabine
• Doxorubicin
• Epirubicin
• Etoposide
• Cisplatin
• Mitoxantrone
Hormones or Anti-Hormones
• Megestrol acetate
• Tamoxifen
• Octreotide
Tyrsine Kinase Inhibitors
• Bevacizumab
• Cediranib
• Cetuximab
• Erlotinib
• Gefitinib
• Lapatinib
• Sorafenib
• Sunitinib
• Vatalanib
Immune Modulators
• Interferon Alpha
• Thymophysin
• Alpha-1-Thymosin
Others
• Bortezomib
• Cox-2-inhibitors
• Everolimus
• Pravastatin
• Rapamycin
• Troglitazone

Table 3. Targeted therapies and others in clinical HCC trials

Name		Target			Study	References
Gefitinib		EGF-R			phase II	(147)
Erlotinib		EGF-R			phase II	(148, 149)
Lapatanib		EGF-R			phase II	(150)
Cetuximab		EGF-R			phase II	(151, 152)
Bevacizumab		VEGF			phase I/II	(153, 154)
Sorafenib		c-Raf1, B-Raf,			phase III	(93)
		PDGF-R, VEGF-R				
Sunitinib		PDGF-R, VEGF-R,			phase II	(155, 156)
		c-KIT, FLT-3				
Vatalanib		PDGF-R, VEGF-R,			phase I	(157)
		c-KIT				
Cediranib		VEGF-R			phase II	(158)
Bortezomib		Proteasome			Phase I/II	(152, 159)

advanced HCC at the time of diagnosis. In this context several nibs and mabs are being evaluated in clinical trials (Table 3). In addition, in experimental preclinical models the mammalian target of rapamycin (mTOR) inhibitors, rapamycin and sirolimus, in combination with bevacizumab and doxorubicin, respectively, were shown to suppress HCC growth (123, 124). Interestingly, everolimus, another mTOR inhibitor, was shown to be effective in patients with metastatic renal cell carcinoma that had progressed on sunitinib/ sorafenib (125).

Apart from exploring novel therapeutic targets, individualization of therapy is of high clinical priority. Tumor-specific mutations/ gene signatures increasingly allow to predict the prognosis and response to therapy in the individual patient. For example, in patients with colorectal cancer it has been recently been shown that cetuximab, is effective only if the tumor carries the ras wild-type gene (126). Thus, it should become possible also in patients with HCC to identify genetic markers that allow to predict whether a given drug will be effective. Genetic marker-based patient selection would improve the drug's overall efficacy and at the same time reduce side effects and costs.

5.4. Experimental strategies

In view of the limited therapeutic options for advanced HCCs a number of experimental strategies are being evaluated, incl. gene and immune therapies (Figure 3) based on suicide, cytokine and antiangiogenic genes or DNA vaccination with tumor-specific genes (127-131), oncolytic viruses (132, 133) as well as novel drugs, e. g., 3-
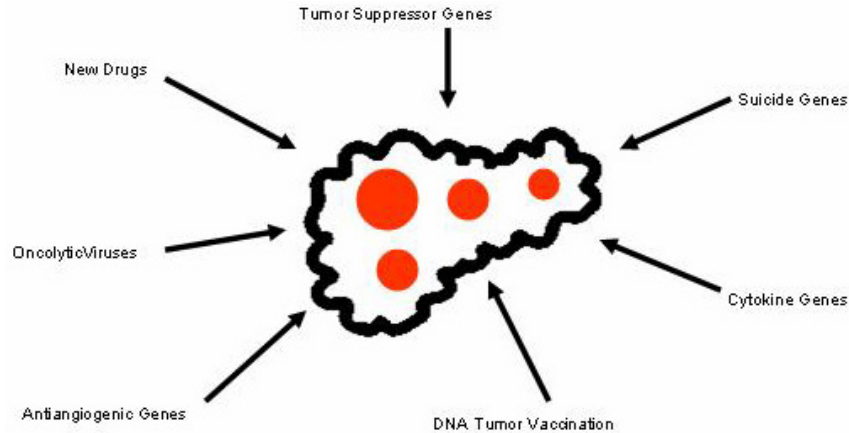
Figure 3. HCC: Experimental strategies.

bromopyruvate (134, 135).

6. HCC PREVENTION

HCC prevention falls into two categories. Primary prevention is aimed at the interference with HCC development at four stages (Figure 4). Stage 1: Interventions at this step are aimed at the prevention of acquired liver diseases. Apart from avoiding liver toxins, including alcohol and certain drugs, or infections with HBV or HCV by hygienic measures, avoiding parenteral exposure to blood, blood products or contaminated needles etc. a prime example is vaccination against HBV infection using the commercially available active and passive vaccines. Several HBV vaccines using natural or recombinant hepatitis B surface antigen (HBsAg) from different sources are well introduced in clinical practice and universal vaccination in Taiwan has indeed already resulted in a decline of the HCC incidence (136). For the prevention of HCV infection, however, there is no effective vaccine available to date. Stage 2: Interventions at this step are aimed at the early treatment of acute hepatitis, thereby blocking their transition into a chronic liver disease. Stage 3: Interventions at this step are aimed at the prevention of the progression of chronic hepatitis to liver cirrhosis that carries a high risk for HCC development. This includes the treatment of inherited, cholestatic or autoimmune liver diseases as well as the treatment of chronic viral hepatitis B or C. Reduction of iron overload by phlebotomy, for example, has been shown to stop the progression of hemochromatosis to liver cirrhosis and HCC. Stage 4: Interventions at this step are aimed at interfering with the molecular events leading to HCC development, usually in a cirrhotic liver. These strategies include all treatment modalities detailed above (stage 3) as far as they can be implemented in patients with compensated or decompensated liver cirrhosis. Finally, LTx in patients with liver cirrhosis before HCC development is a highly effective preventive measure.

After successful HCC resection or non-surgical ablation, HCC recurrence in the remaining, in most cases

cirrhotic liver is the major limitation of the life expectancy of these patients. The probability of recurrence is about 50% within 3 years after successful treatment (36, 137). Strategies to prevent HCC recurrence are therefore central to the improvement of survival of HCC patients after initial cure. Apart from LTx after successful resection (48), the strategies explored to date include the administration of polyprenoic acid, an acyclic retinoid (138), of interferon alpha (139) and of interferon beta (140). Further, adoptive immunotherapy (141) and intraarterial injection of ^{131}iodine-labeled lipiodol (142, 143) has been evaluated in clinical studies. Further, postoperative adjuvant TACE may also improve survival in patients with risk factors for residual tumor (91). All these interventions have resulted in lower HCC recurrence rates. These findings have to be confirmed, however, in larger randomized controlled studies demonstrating a clear clinical benefit before secondary prevention with one the strategies mentioned above should enter clinical practice.

In an animal model of chemical hepatocarcinogenesis using dimethylnitrosamine in which only males developed HCCs it has recently been shown that tumor development in males can be prevented by estrogens (144). The mechanism of action was mediated through inhibition of interleukin-6 synthesis in macrophages that was induced by hepatocellular necrosis. While these findings may in part explain the predominance of male patients with HCC, the data need to be reproduced and extended in order to demonstrate their clinical relevance with respect to a preventive strategy in patients with chronic liver diseases. Further, recent experimental data suggest that c-kit inhibition by imatinib mesylate attenuates progenitor cell expansion and prevents HCC formation in mice while it is not effective as a therapeutic agent (145, 146).

7. SUMMARY AND PERSPECTIVES

HCC is one of the most common malignant tumors in some areas of the world with an extremely poor prognosis. HCC treatment is based on randomized
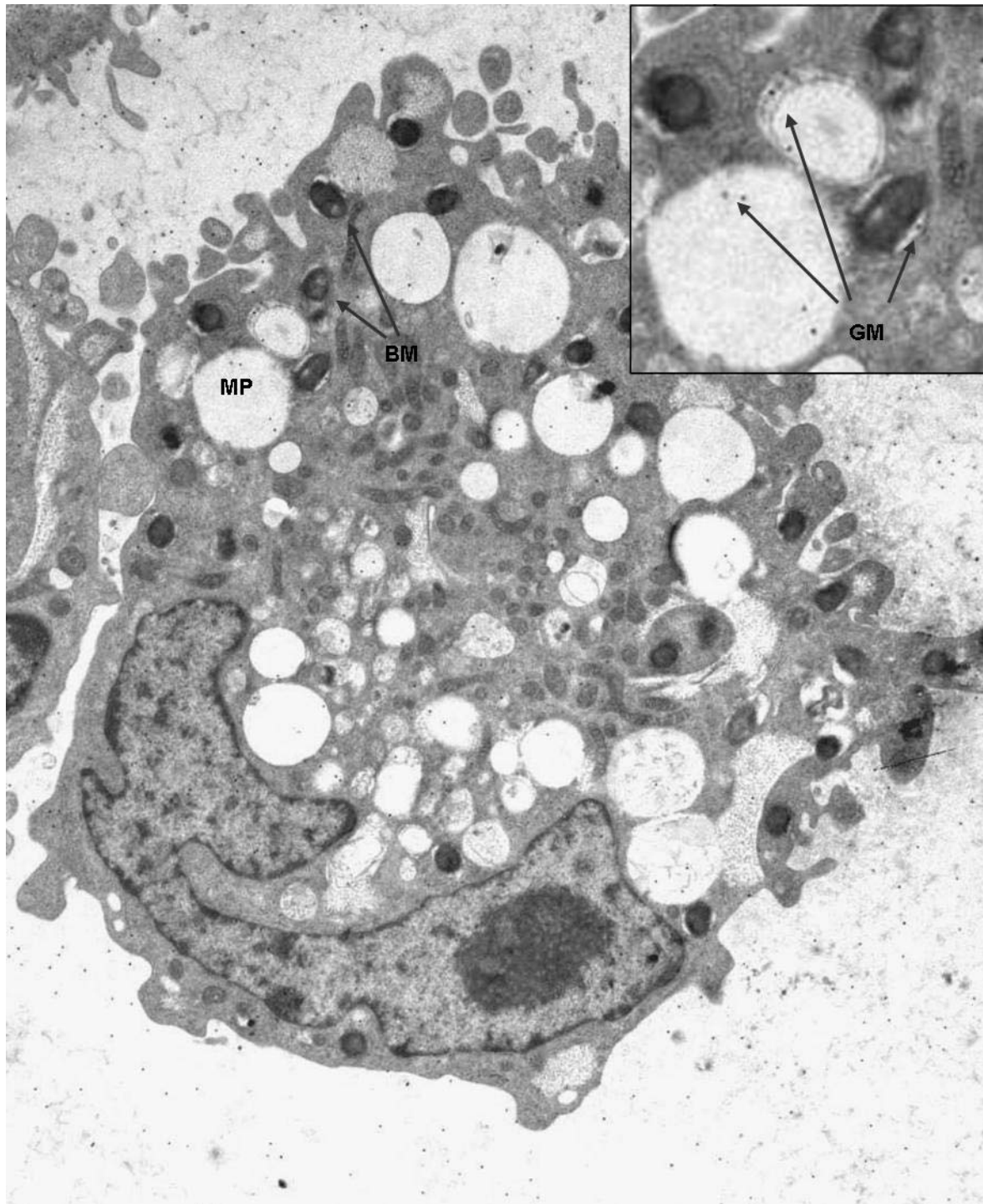
Figure 4. Primary HCC prevention. 1: prevention of liver disease, e.g., by hygienic measures, prevention of exposure or vaccination against HBV infection, abstinence from alcohol etc. 2: prevention of chronic hepatitis; e.g., by treatment of acute hepatitis C, abstinence from alcohol, treatment of hereditary liver diseases etc. 3: prevention of liver cirrhosis, e.g., by antiviral treatment of chronic hepatitis B or C, treatment of other chronic liver diseases. 4: prevention of HCC development in liver cirrhosis, e.g., by antiviral treatment, inhibition of fibrosis, liver transplantation, etc.

controlled trials and many observational studies. Treatment options fall into five main categories: (1) surgical interventions, incl. tumor resection and LTx, (2) percutaneous interventions, incl. PEI and RFA, (3) transarterial interventions, incl. TAC, TAE and TACE as well as the combination of TACE and RFA and (4)

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radiotherapy, incl. external radiation and SIRT and (5) drugs as well as gene and immune therapies. While surgery and percutaneous as well as transarterial interventions are effective in patients with limited disease (up to 3 lesions <3 cm in diameter or 1 lesion <5 cm in diameter) and compensated underlying liver disease (cirrhosis Child A), at the time of diagnosis more than 80% of the patients present with multicentric HCC and advanced liver disease or comorbidities that restrict the therapeutic measures to sorafenib or best supportive care.

In order to reduce morbidity and mortality from HCC, therefore, early diagnosis and the development of novel systemic therapies for advanced disease, incl. drugs, gene and immune therapies as well as primary HCC prevention are of paramount importance. Further, secondary HCC prevention after successful therapeutic interventions needs to be improved in order to make an impact on the survival of patients with HCC. New technologies, including gene expression profiling and proteomic analyses, should allow to further elucidate the molecular events underlying HCC development and to identify novel diagnostic markers as well as therapeutic and preventive targets.

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