肝病的精准医学研究与应用

解放军总医院第五医学中心

国家感染性疾病临床研究中心

王福生

提纲

一、背景

二、肝脏免疫特点

三、慢性乙肝、艾滋病精准治疗？

四、前景与展望

提纲

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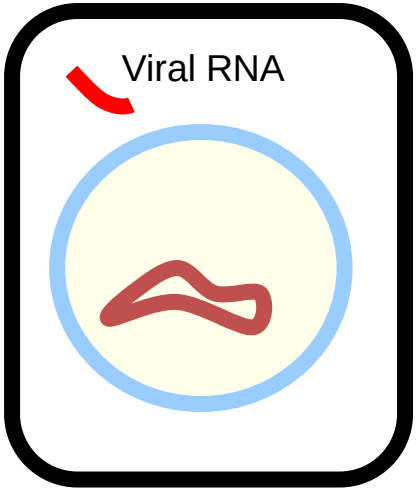
Three chronic viral diseases: CURE?

HCV



TREATMENT

No viral reservoir

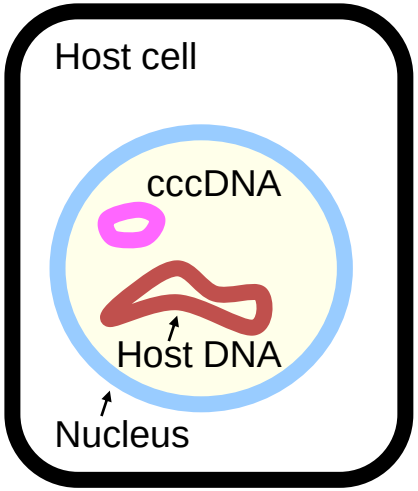
↓

Combination therapy

↓

DAAs: CURE!!!

HBV



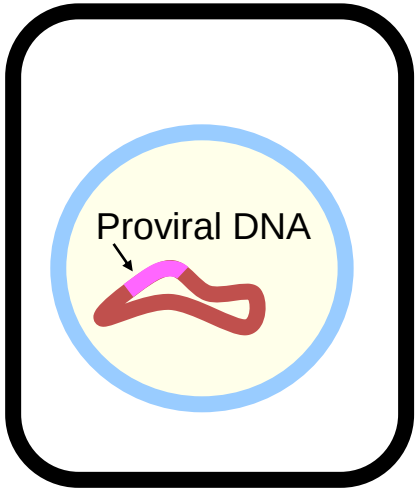
TREATMENT

cccDNA (viral reservoir)

↓

Massive HBsAg

HIV



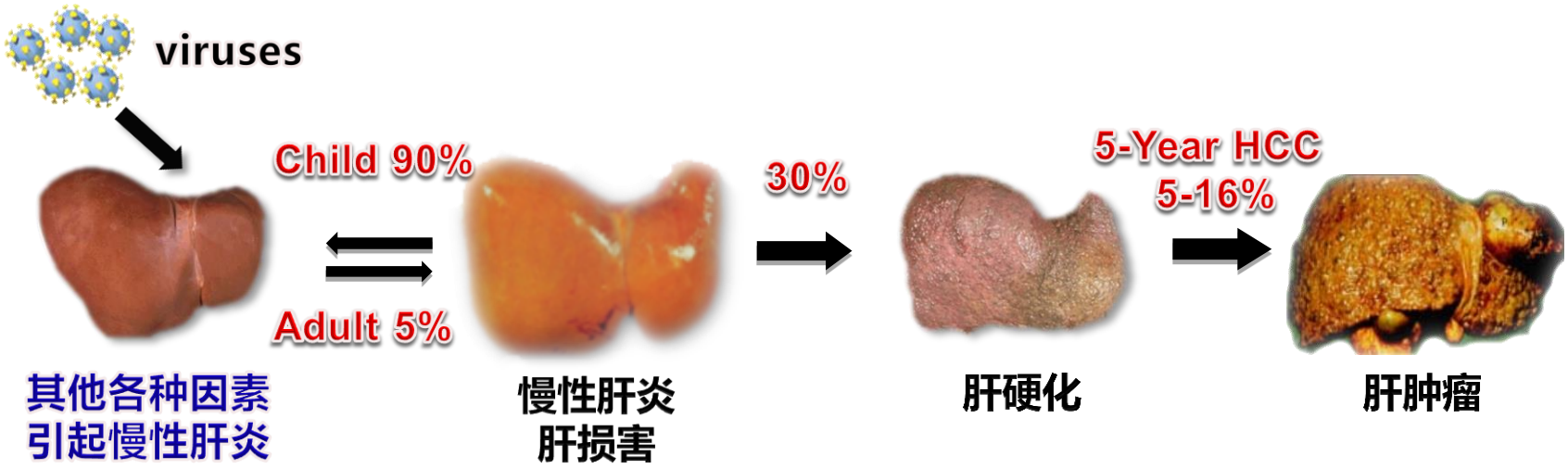
TREATMENT

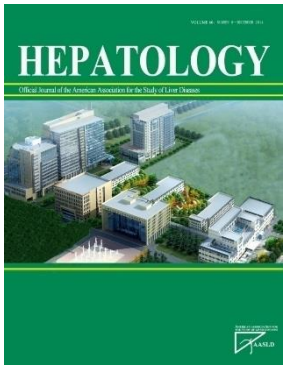
↓

HIV DNA (viral reservoir)

Few chronic cases are with self-functional cure!

我国是肝病大国，慢性乙肝危害严重





我国肝病数据：9000万 HBV、HCV肝炎

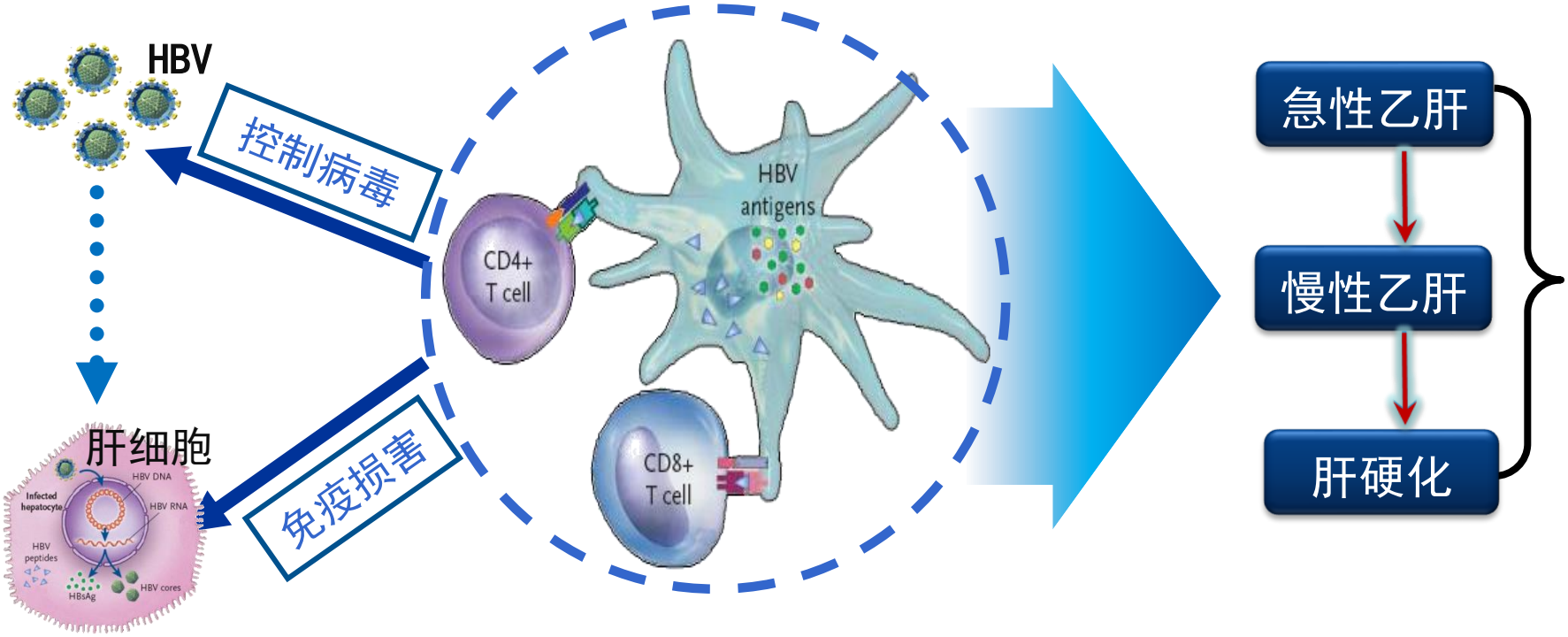
2亿其他肝病：ALD、DILI、AID等

Wang FS, Fan JG, Zhang Z, et al. Hepatology 2014

慢性乙型肝炎防治指南（2015更新版）. 中华肝脏病杂志, 2015, 23.

免疫应答是控制疾病进展的关键

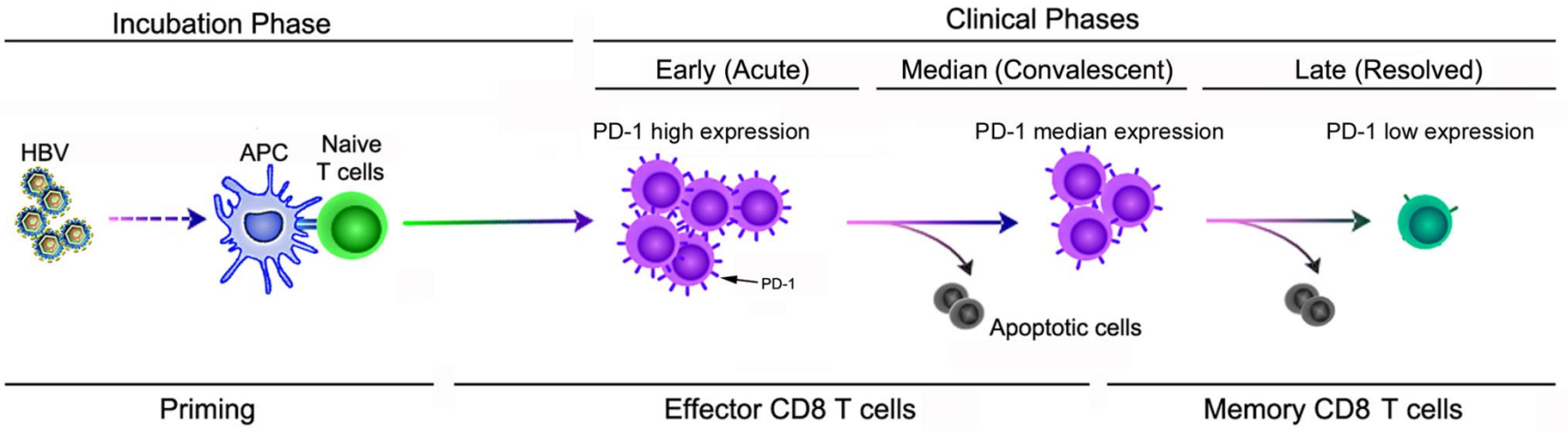
病毒感染 → HBV特异性免疫应答 → 疾病进展



王福生. *Hepatol Res* 2007； *Exp Rev Gastro Hepatol* 2009

急性乙肝：PD-1有效调控免疫应答

HBV特异性CD8 T细胞

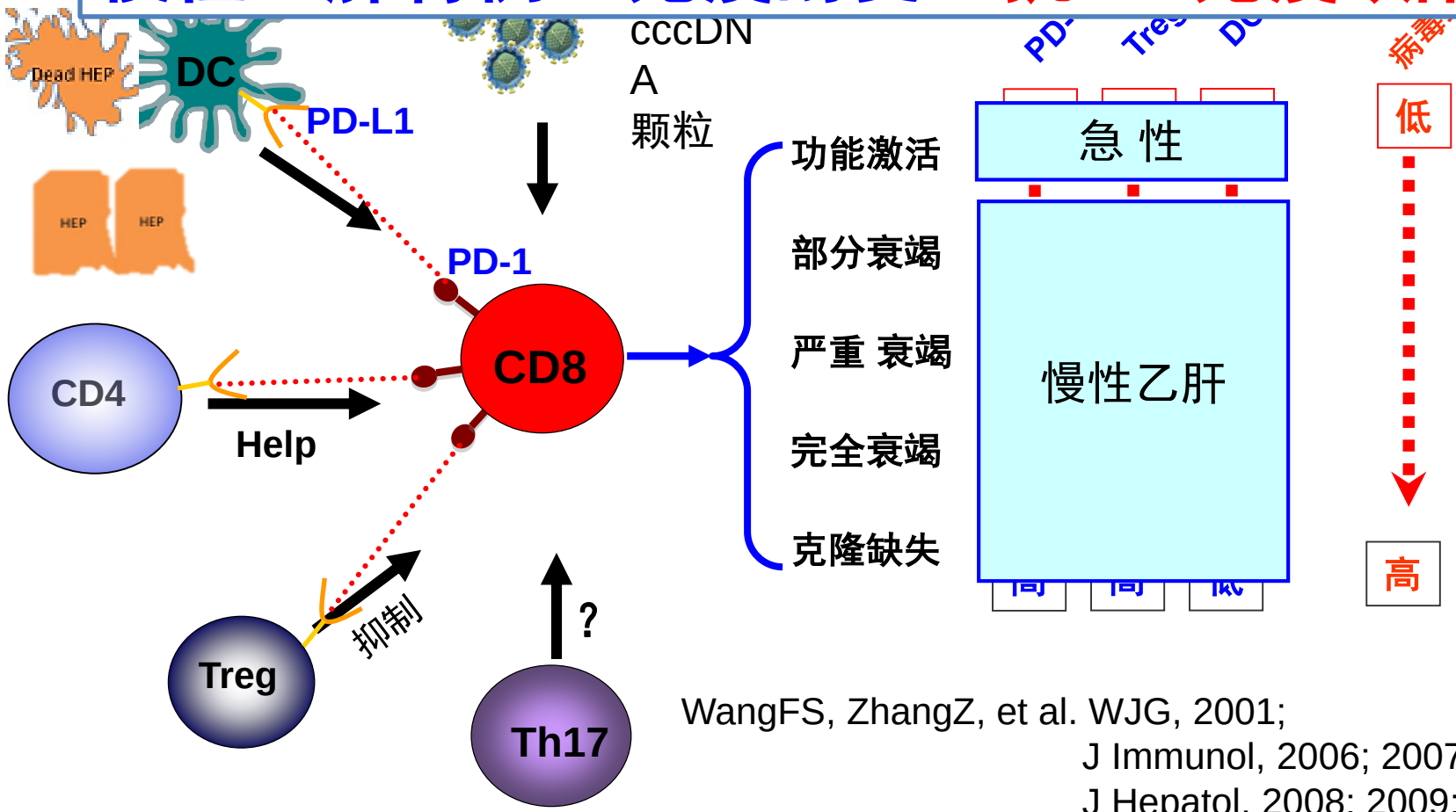


Zhang Z, et al. J Hepatol, 2009

慢性乙肝：HBV特异性T细胞功能损伤

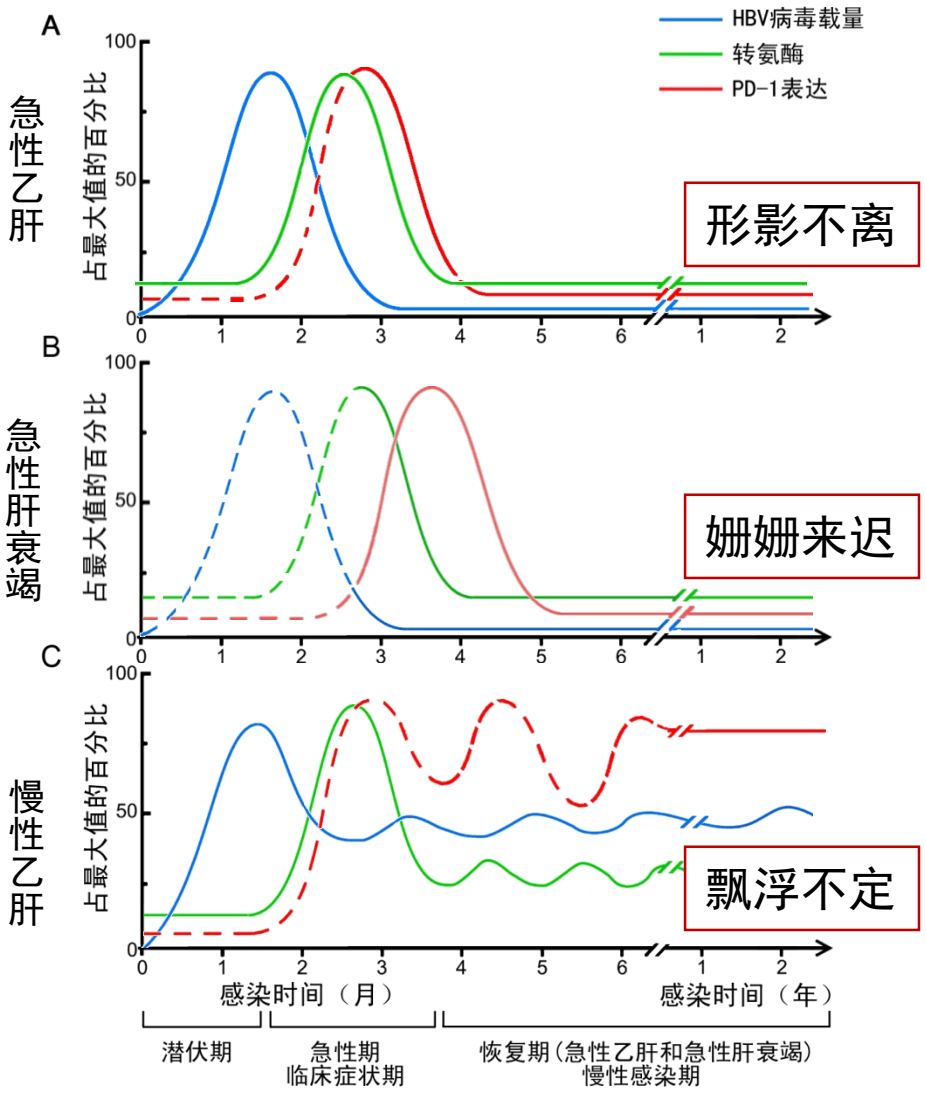
——Treg细胞、DC和PD-1等发挥作用

慢性乙肝特例：免疫耐受（抗HBV免疫缺陷）



WangFS, ZhangZ, et al. WJG, 2001;
J Immunol, 2006; 2007; 2008
J Hepatol, 2008; 2009;
Clin Immunol, 2007; 2008

PD-1表达变化预测HBV急性感染临床转归



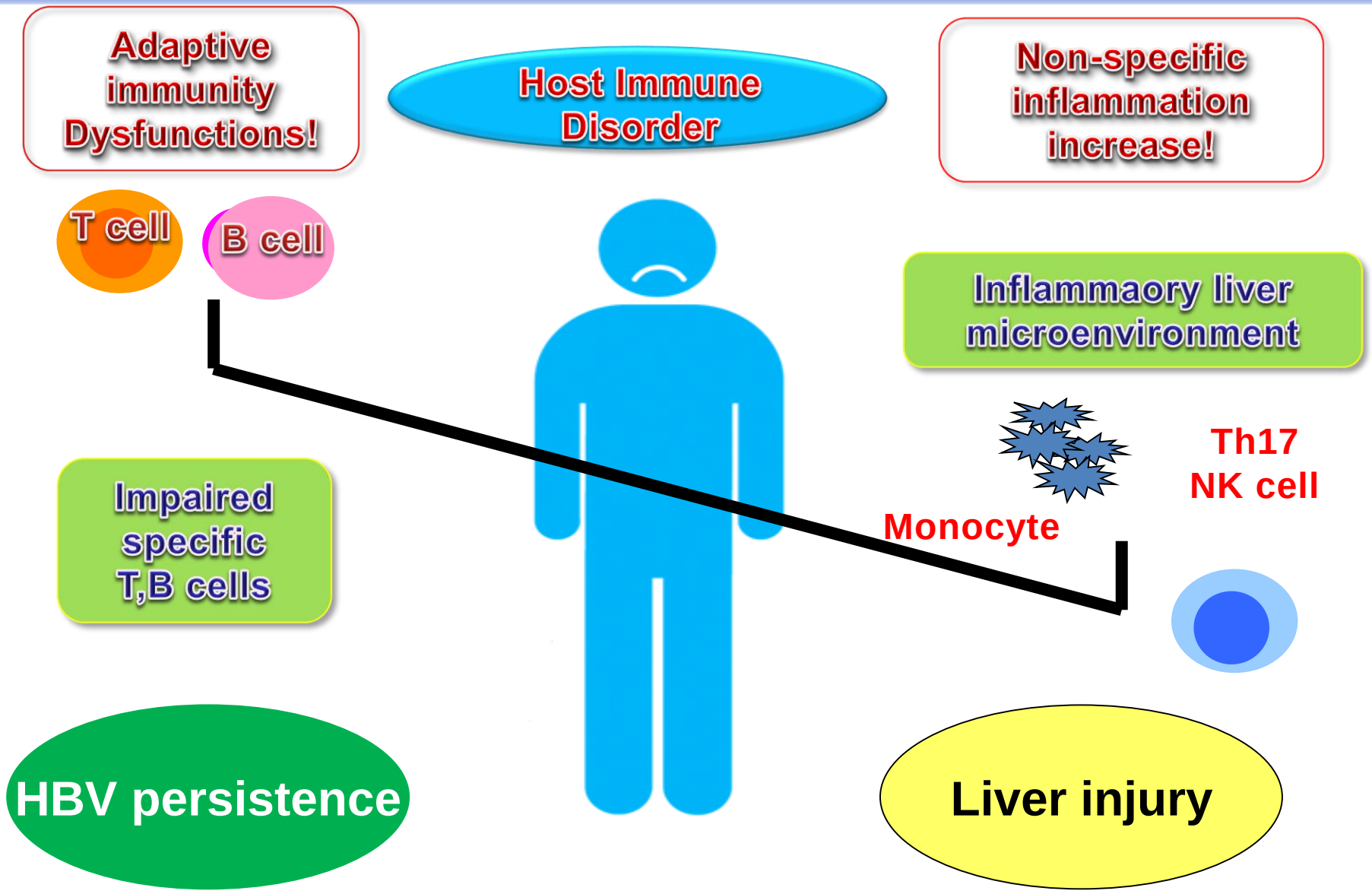
国际权威Ahmed教授等撰写了4页评论（同期发表）——

该文通过深入研究PD-1在HBV感染中的作用，发现PD-1表达水平与临床转归直接相关，并阐明了肝内作用的机制 - - - - - -

for the immune response in the human liver to HBV and HCV infections.[6,7] The articles by Zhang et al and Nakamoto et al further our understanding of the role of PD-1 in HBV and HCV infections, respectively, by emphasizing clinical outcomes in patients in relation to the level of PD-1 expression and importantly by focusing on intrahepatic immune responses.[6,7] Studies of antigen-specific $CD8^+$ T cells from the peripheral blood of patients with HBV and HCV infections have previously indicated that the PD-1/PD-L1 system plays an important role in the PD-L1 in the liver during the acute and chronic phases of infection, Zhang et al report that more $CD4^+$ and $CD8^+$ T cells in the liver expressed PD-1 during the acute phase in comparison with the chronic phase, and more in the chronic phase than in healthy liver biopsies. Furthermore, PD-L1 expression was found on Kupffer cells, sinusoidal epithelial cells, and $CD11c^+$ dendritic cells, sim-

Zhang JY, et al. Gastroenterology 2008

The immuno-pathological characteristics of CHB



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Immune responses in the liver are biased toward tolerance

Induction of Immunological Tolerance by Porcine Liver Allografts

by
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Liver allografting experiments with pigs show that animals given no immunosuppression will survive for prolonged periods with orthotopic liver transplants. Similar animals can reject skin, kidneys and hearts rapidly. Orthotopic and accessory heterotopic liver allografts protect preferentially from rejection grafts of donor specific skin, kidney and possibly heart. Injected soluble liver antigen may also protect donor specific tissue from rejection. It is suggested that allogeneic liver can induce immunological tolerance in immunologically mature pigs.

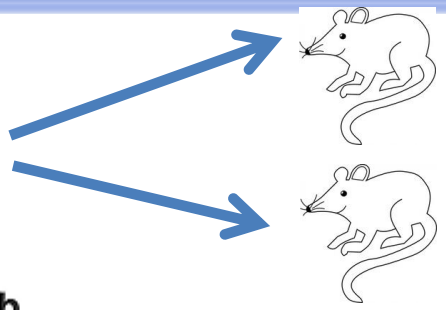
Question:

What is the mechanism of the liver tolerance?

Is this kind of tolerance relevant to liver disease?

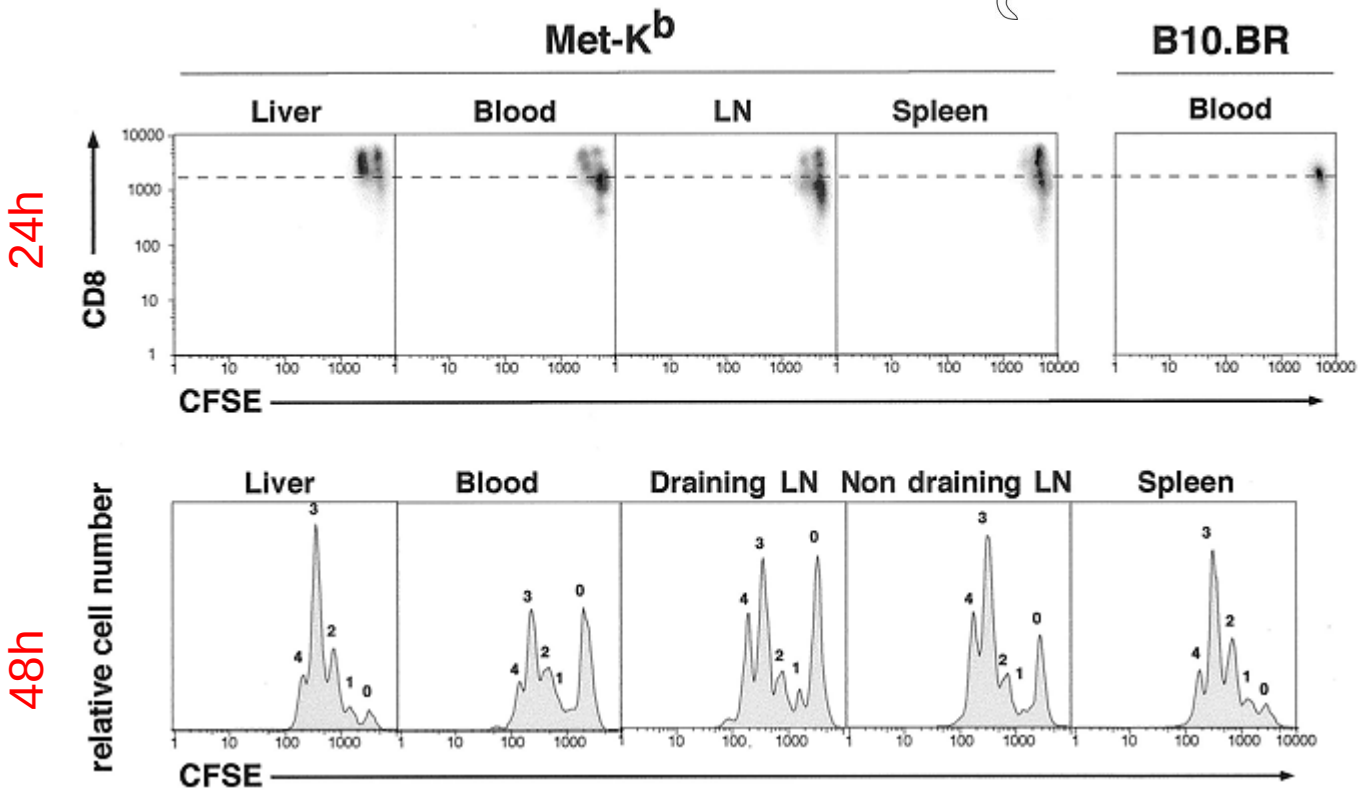
First: the liver act as secondary lymphoid organ

Naive Des-TCR transgenic
CD8 T cells specific for H-2K^b



B10.BR

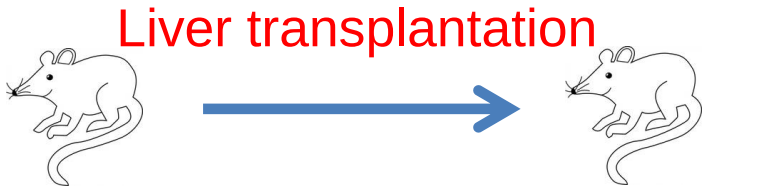
Met-K^b mice expressing
H-2K^b in the liver

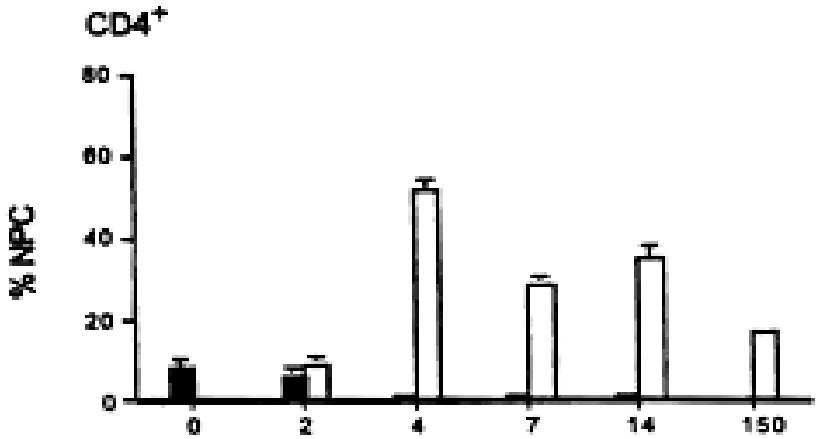


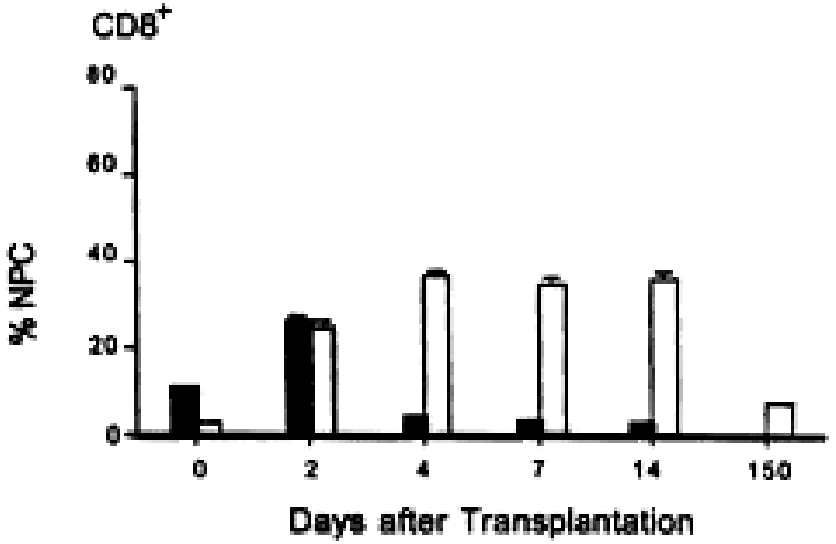
Naïve CD8 T cells specific for liver antigens are activated locally in the liver

Bertolino P, et al.J Immunol. 2001;166(9):5430-8.

Apoptosis within spontaneously accepted mouse liver allografts



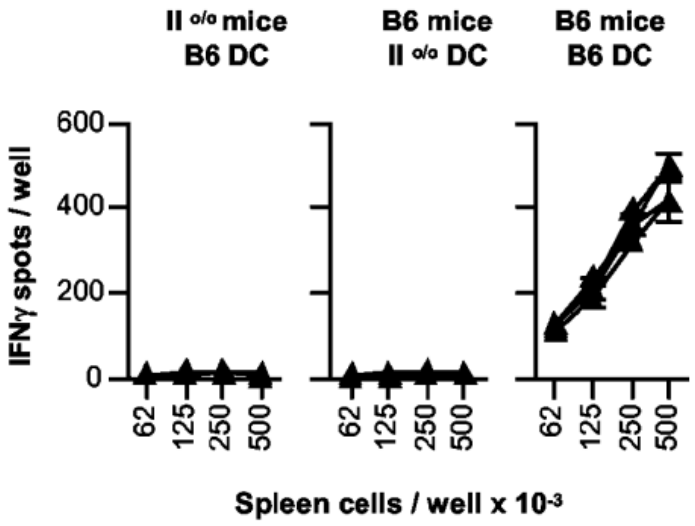


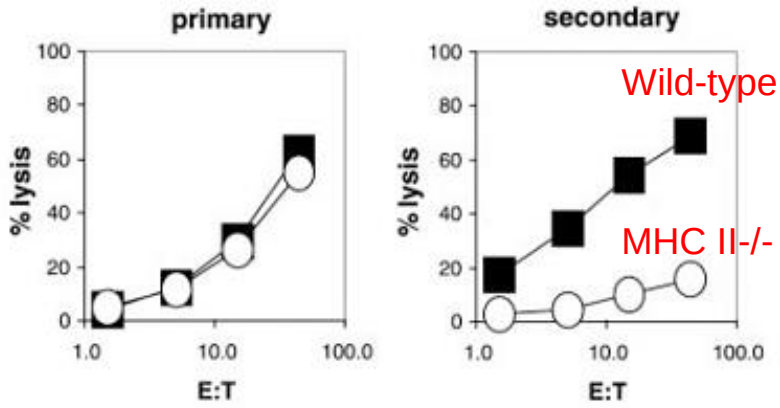


Donor T cells within the liver were rapidly replaced within 2 to 4 days of transplantation with those of the recipient

Qian S, Lu L,et al. J Immunol. 1997;158(10):4654-61.

Fundamental theory





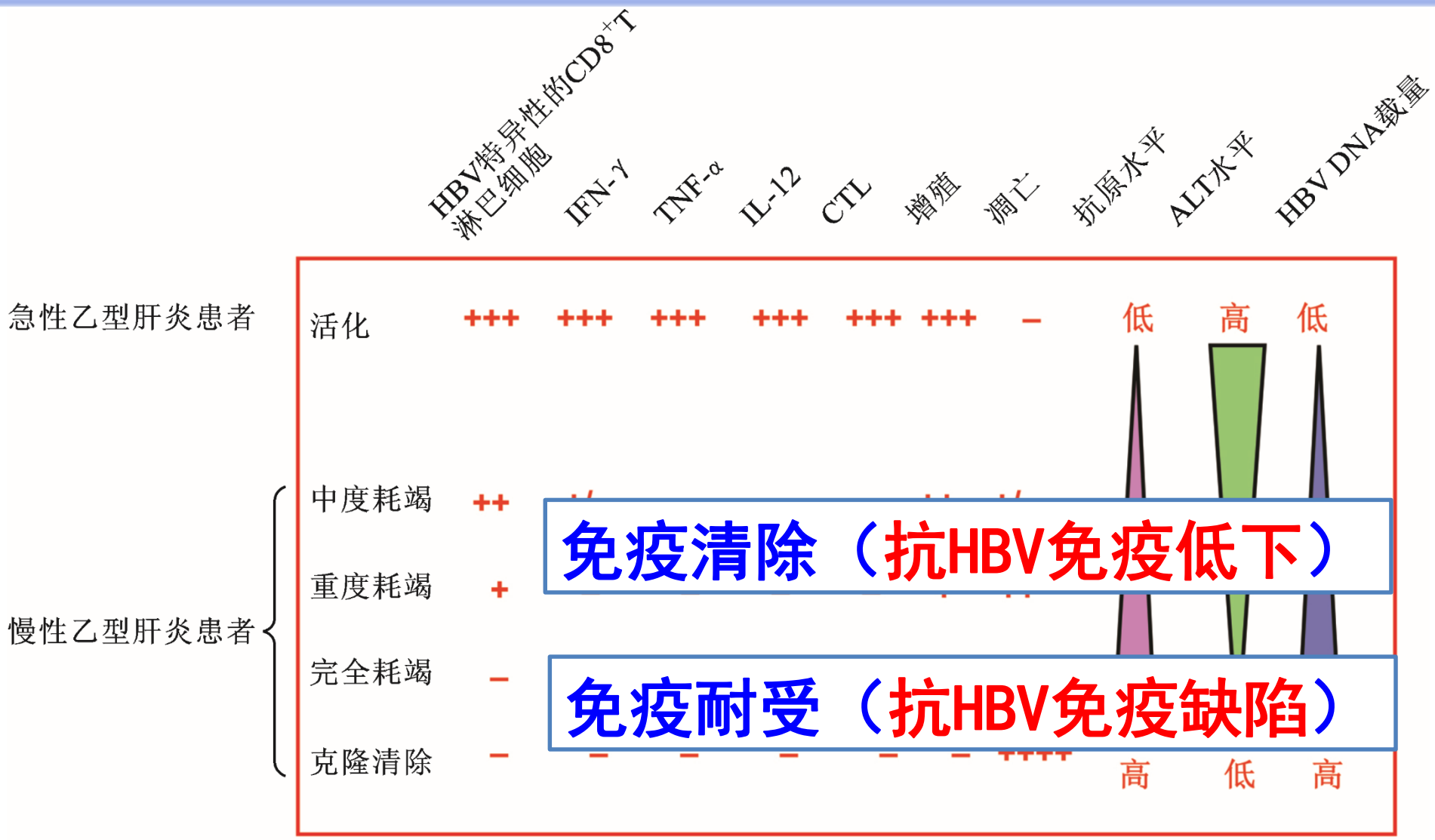
CD4+ T cell help can be essential for primary CD8+ T cell responses in vivo.

Defective CD8 T cell memory following acute infection without CD4 T cell help.

Wang JC, et al. J Immunol. 2003;171(12):6339-43.

Sun JC, et al. Science. 2003;300(5617):339-42.

Relevant to liver disease : evidence from CHB



Expert Rev Gastroenterol Hepatol 2009;3(5):499-512.

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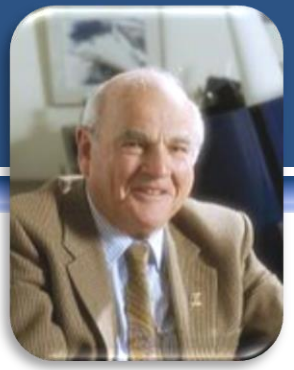
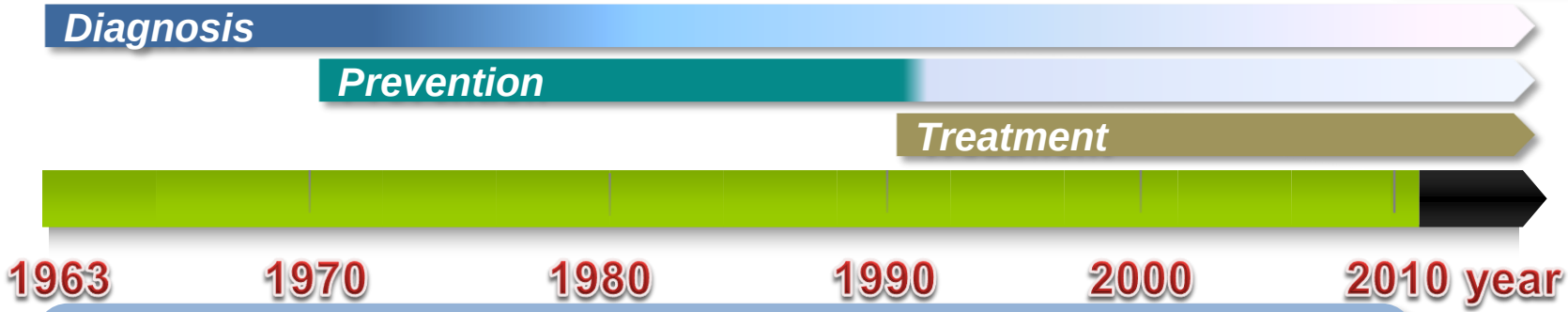
Discovery of HBsAg: a milestone!



Dr. BS. Blumberg: Nobel laureate in 1976



- 1963: Discovery of "Au" Ag
- 1967: Possible association between "Au" Ag and hepatitis B
- 1970: Au Ag screening in blood donors by WHO

Immune Related

- 1971: Finding HBsAg vaccine
- 1981: Approval of HBV vaccine (FDA)
- 1985: Approval of HBV vaccine in China
- 1991: HBV vaccination by WHO

- 1992: Approval of IFN-α
- 1998: Approval of first oral antiviral drug--Lamivudine
- 2000: PEG-IFN- α

乙型肝炎临床检测指标：诊治、预后

1. 病毒学：病毒载量、基因型、耐药变异、准种

病毒库（cccDNA)、HBV RNA

2.乙肝五项：抗原抗体两对半（病毒和免疫指标）

HBsAg、HBeAg、

HBsAb；HBeAb；HBcAb

3. 免疫学指标：HBV特异性CTL、其他指标

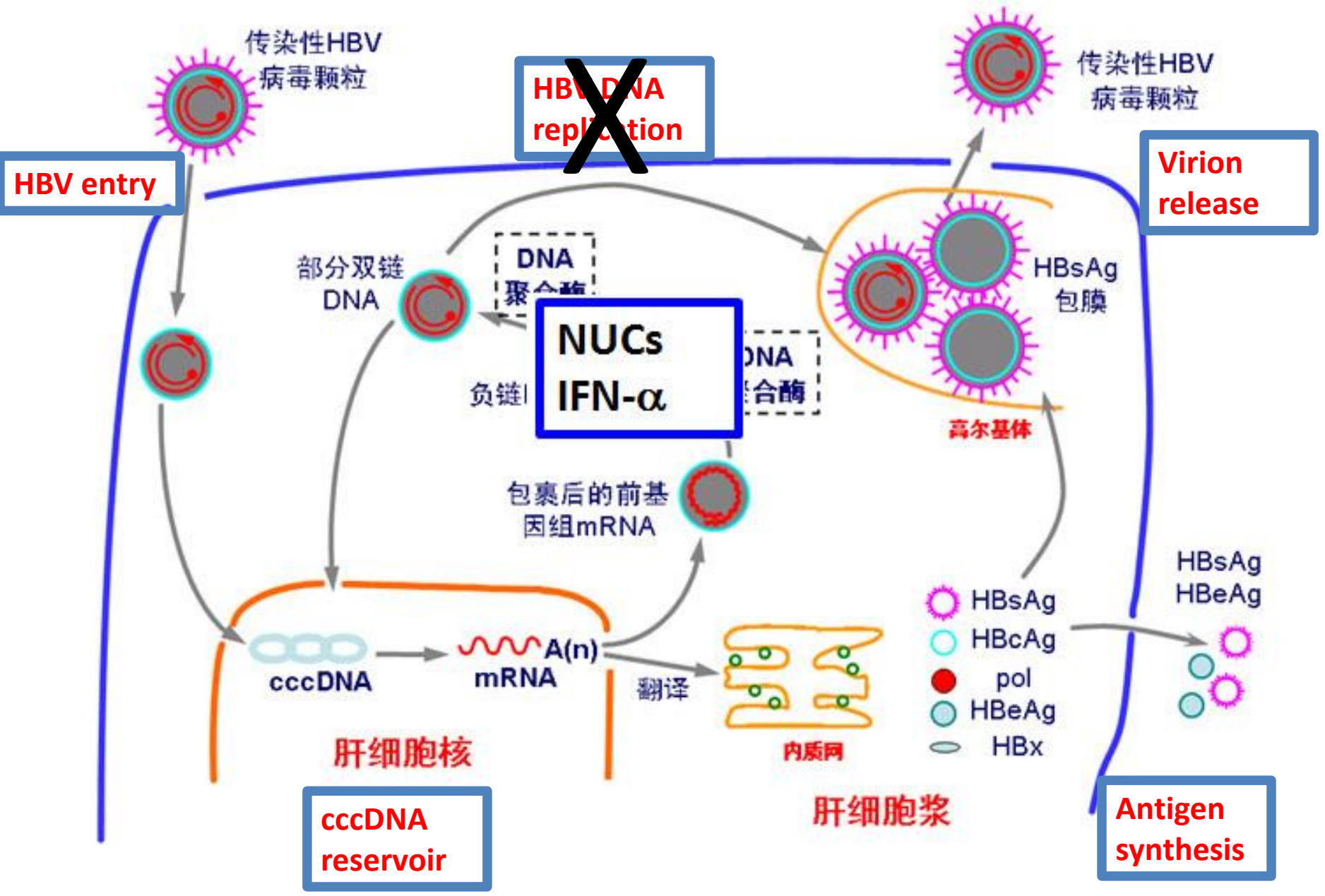
Efficacy of currently used drugs for two years.

Treatment	Undetectable HBV DNA in HBeAg(+) patients	Anti-HBeAg serconversion in HBeAg(+) patients	HBV DNA < 300/400 copies/ml in HBeAg (-) patients
Peg-interferon	25	30	63
Lamivudine	39	22	72
Adefovir	21	12	51
Entecavir	67	22	90
Telbivudine	60	26	88
Tenofovir disoproxil fumarate	74	21	91

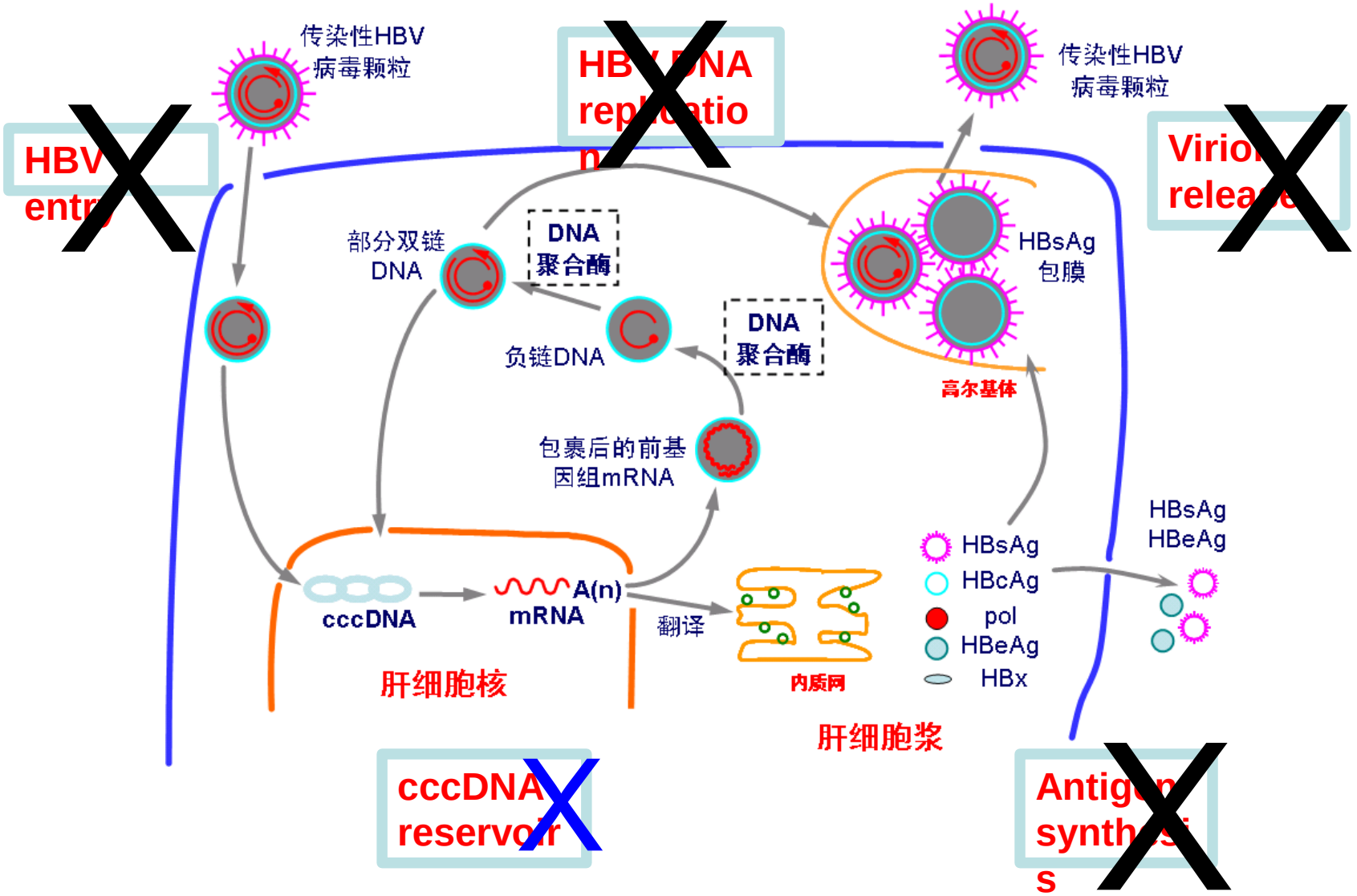
TAF will be available in near future!

Manzoor S, et al. World J Gastroenterol. 2015;21(44):12558-75.

Efficacy of first-line drugs is limited against HBV



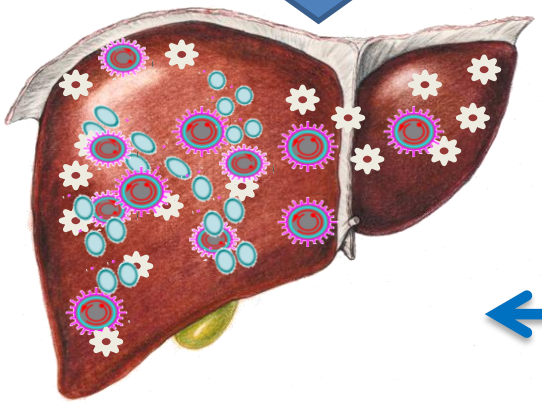
免疫发挥清除或控制病毒（乙肝自愈）



Previous immune therapy is not successful!

Antiviral drugs
- NUCs
- Pegylated – IFN-α

ART



Critical concerns for immune therapy

- Ag-Ab immune complex Therapeutic vaccine
- HBV DNA Vaccine
- Immunomodulating cytokines (IL-2,IL-12)

- No full success for a single immune therapy in clinical trial so far !

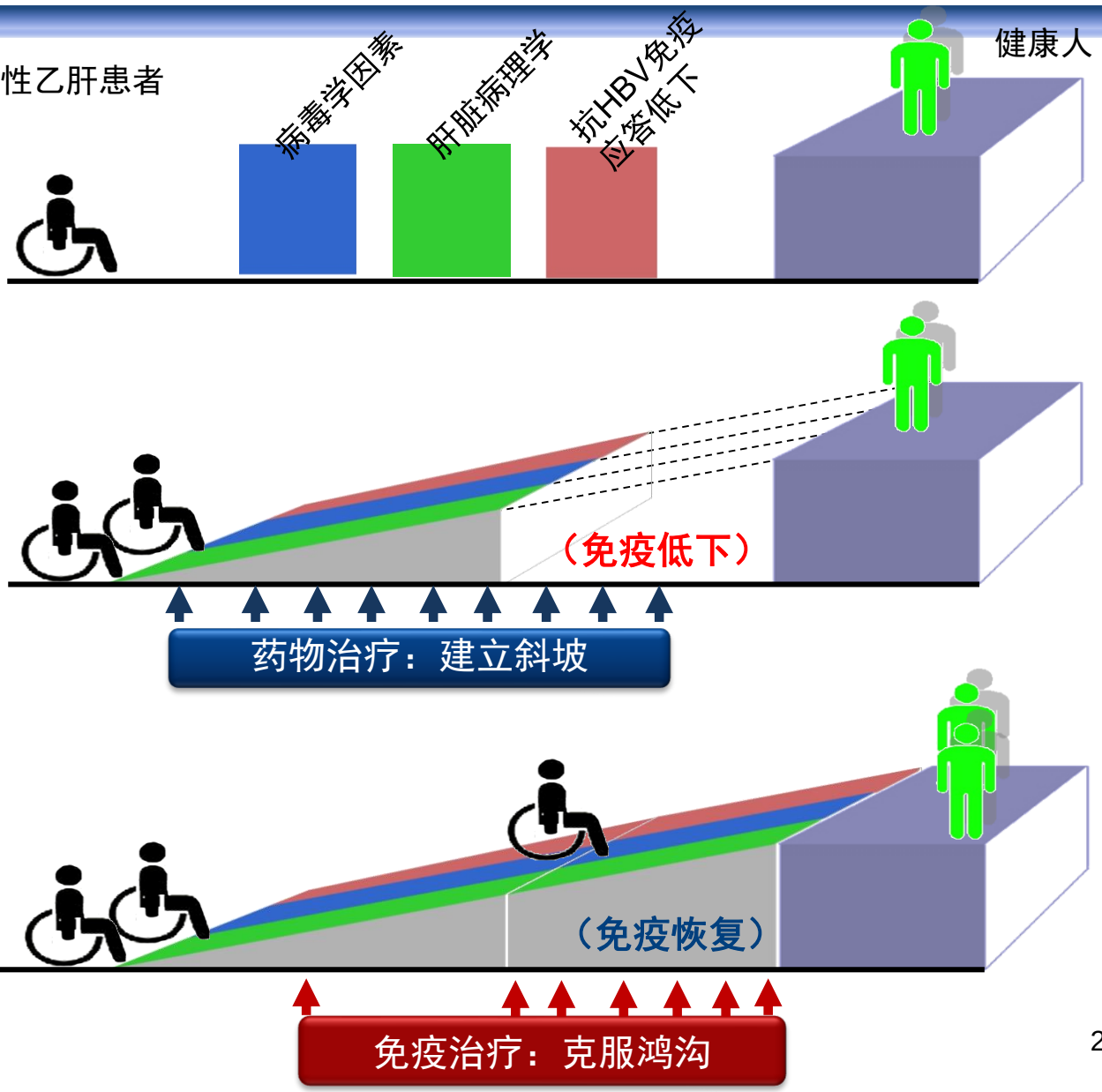
HBV virions

HBeAg

HBsAg

首先提出了抗病毒治疗的“爬坡假说”

构建斜坡、克服鸿沟



王福生 等.
Expert Review Gastro Hepatol 2009
中华肝脏病杂志 2009

慢性乙肝的关键免疫学机制

慢性乙肝

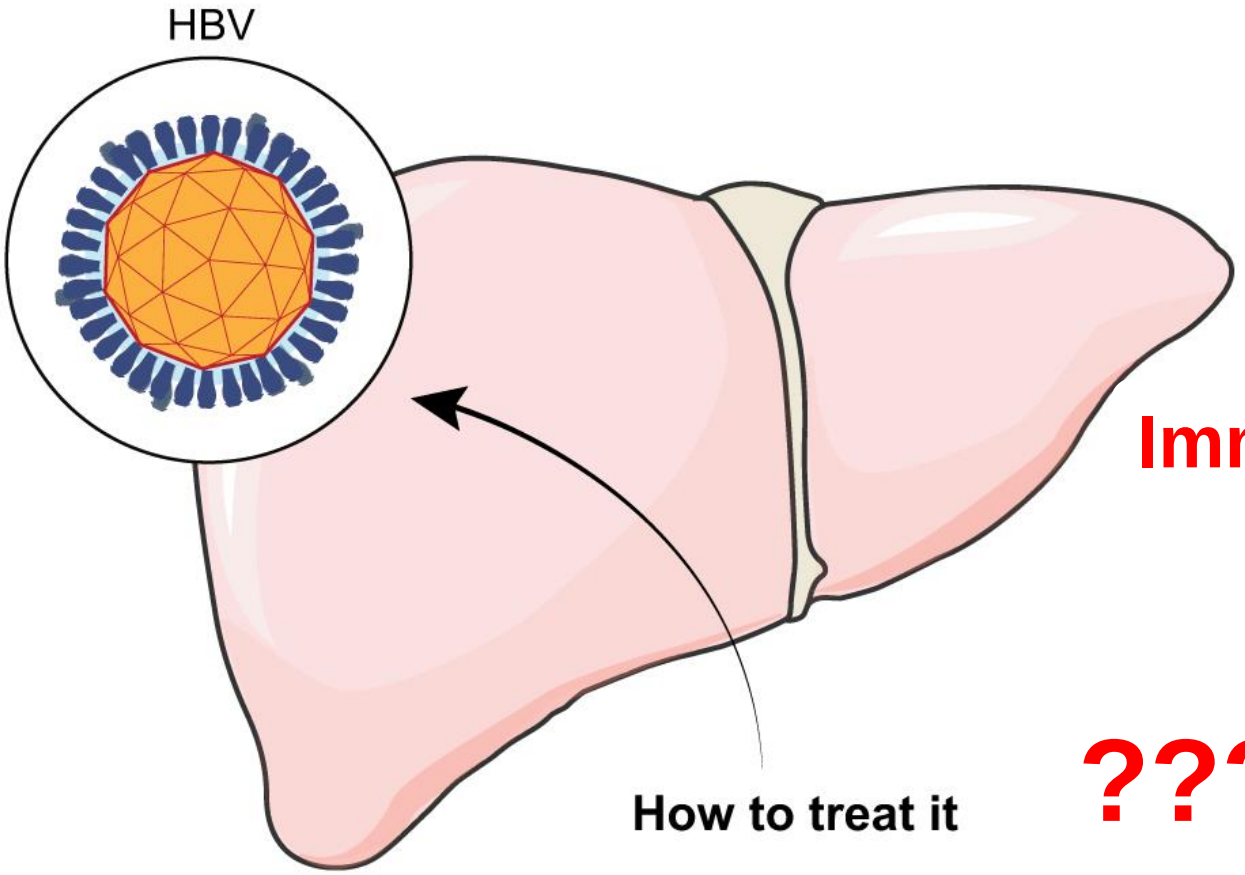
核心问题

特异性CD8细胞功能低下？

肝脏、慢乙肝免疫学机制？

加大临床和转化医学、基础医学的研究！！！

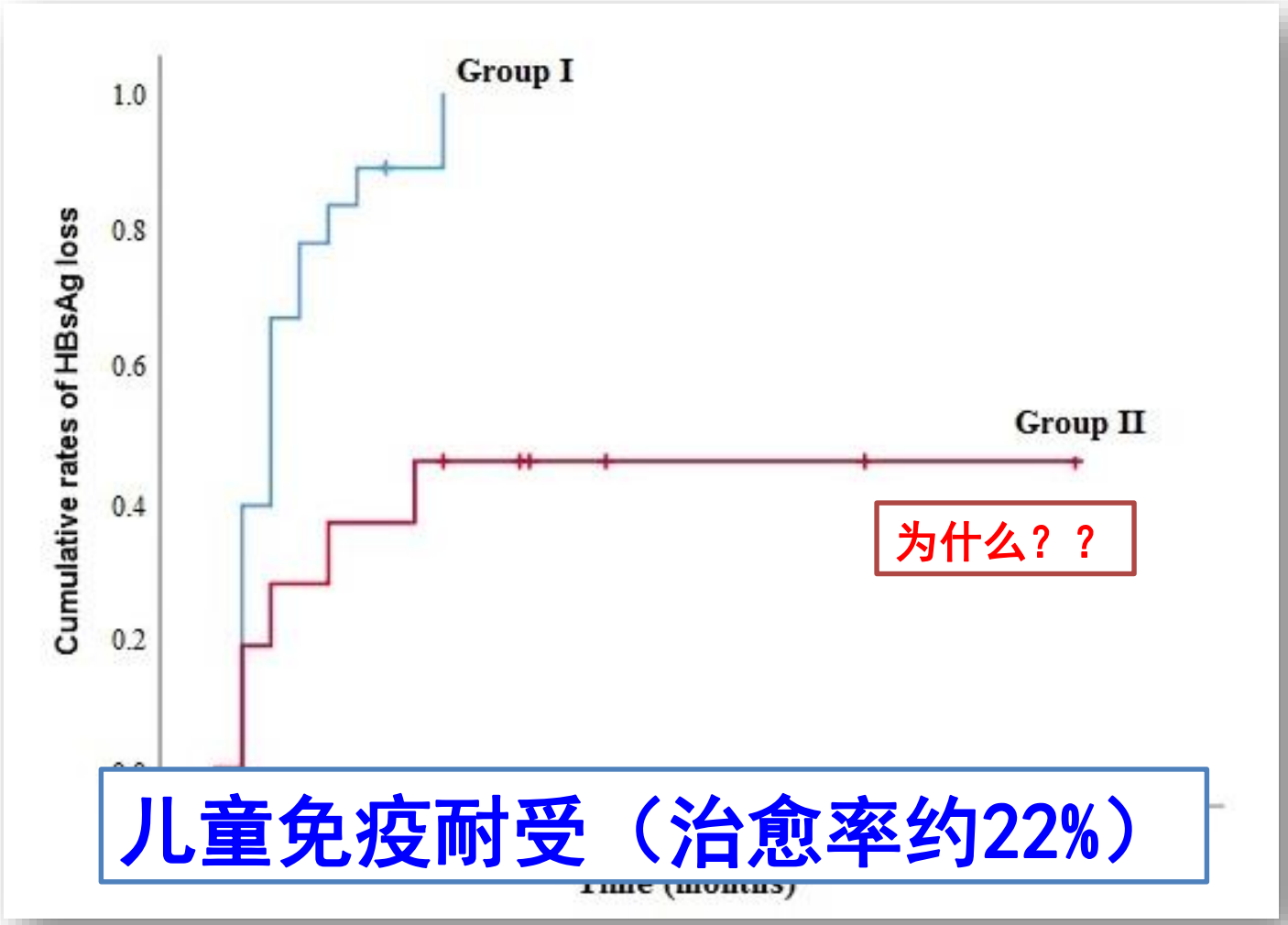
免疫清除期儿童慢性乙肝的治疗



Immune-active liver

???

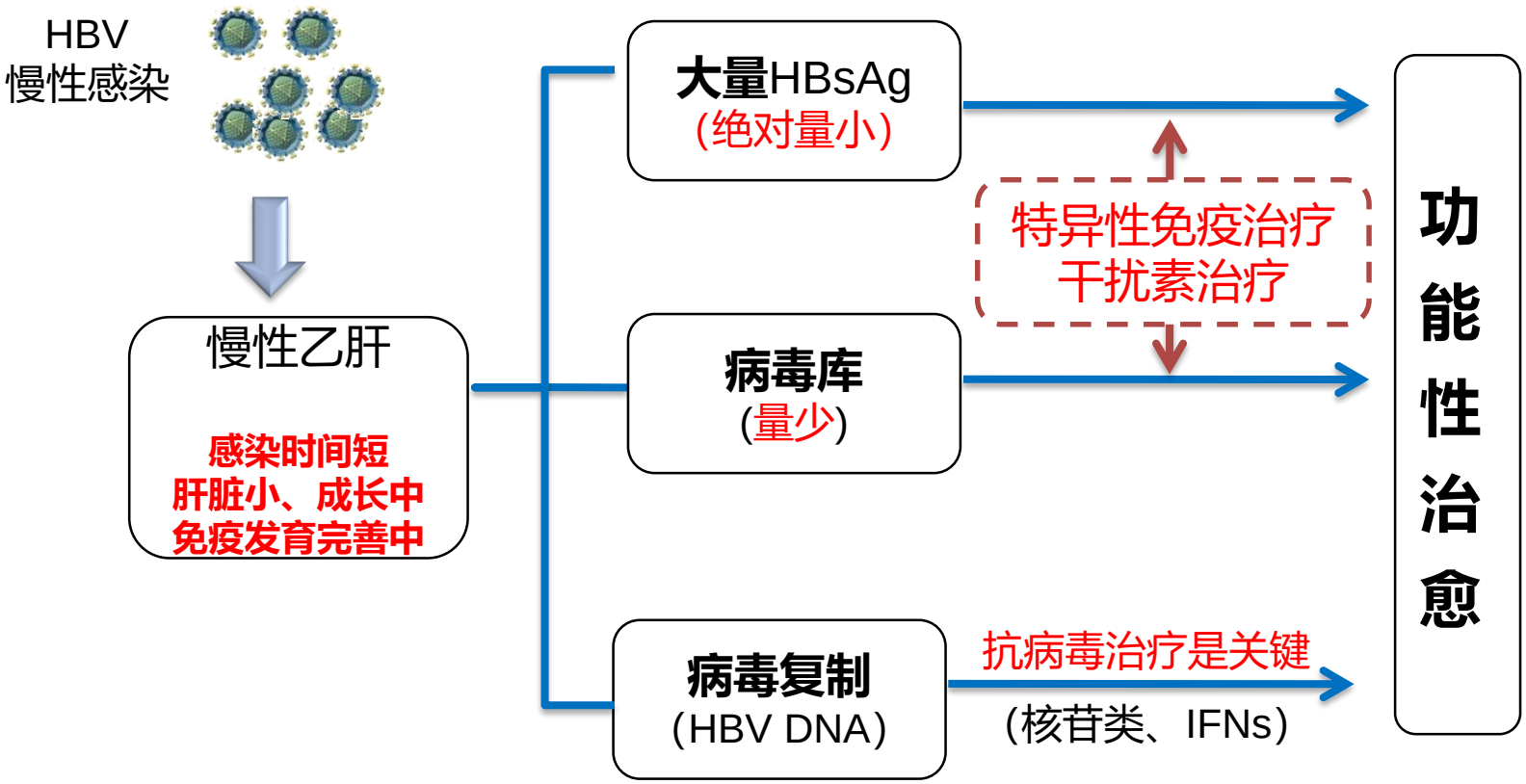
治疗随访期间HBsAg累积清除率



Zhu S, et al. J Hepatol. 2019, online publication

治愈儿童慢性乙肝的关键问题

Cure of CHB children: Critical issues



2018年诺贝尔奖获得者

詹姆斯·艾利森教授

本庶佑教授

?

临床免疫治疗发展新机遇

药物制剂的三个发展阶段

传统片剂、胶囊、
注射制剂

单克隆抗体、基因工程抗体、
重组疫苗

CAR-T、TCR-T、AACT

化合物药物 → 蛋白质药物 → 细胞药物

广谱治疗

靶向治疗

个体化治疗

新技术具有引领性、突破性、颠覆性；

临床诊疗更及时、更准确、更智能！

重大疾病的治疗有更多、更好的选择！

艾滋病能够获得治愈吗？

能够！

目前治愈的两个HIV病人：柏林病人和伦敦病人

CCR5△32$^{-/-}$

骨髓移植

功能性治愈

迄今9年均检测不到HIV

Hutter G et al, N Engl J Med. 2009

Nature 2019

同种异体过继免疫细胞治疗

Allogenic Adoptive Immune Therapy （AAIT）

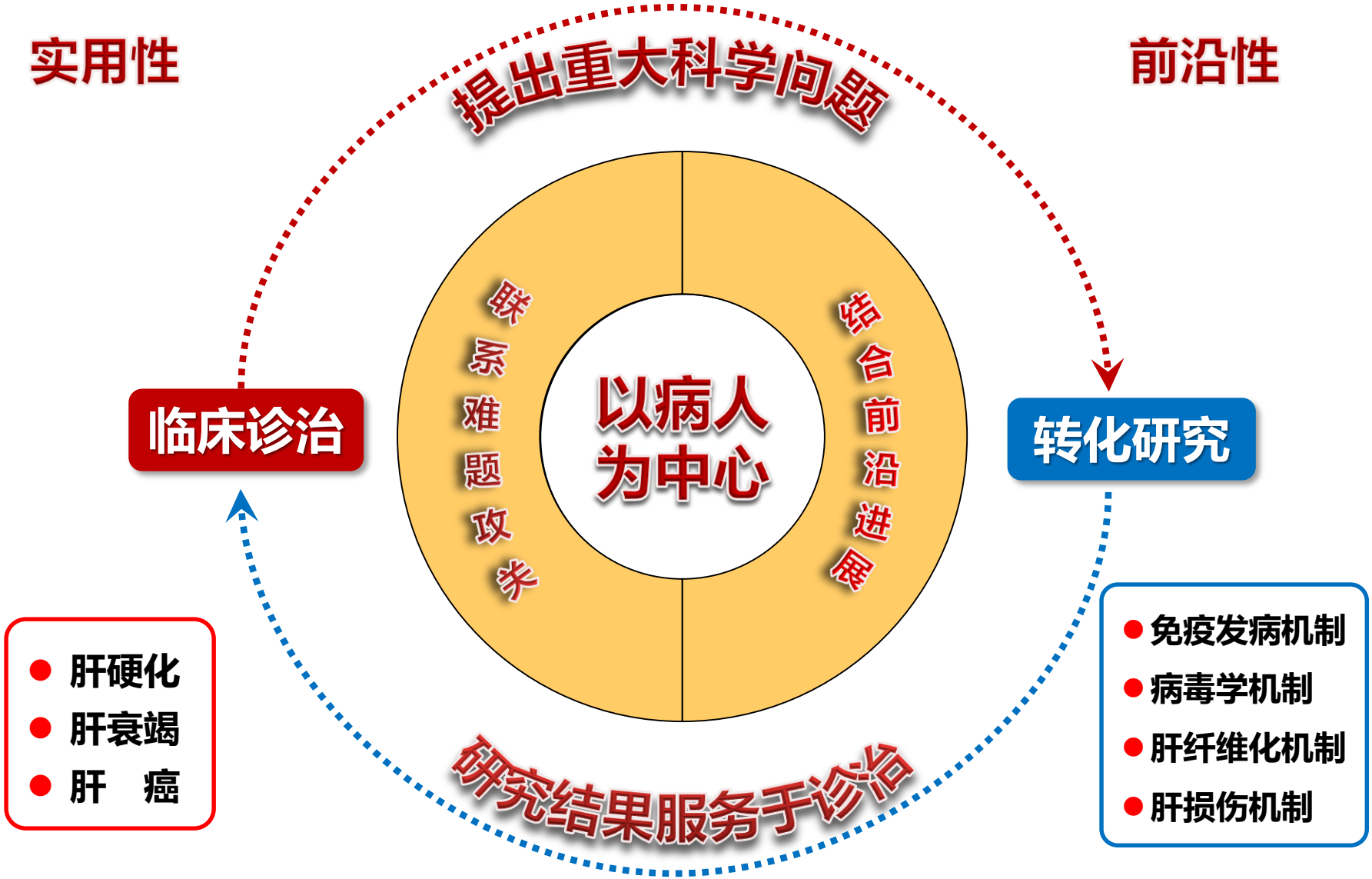
难治性艾滋病（极晚期CD4 <50/μL）特点：

HIV活跃
持久复制 → 大量破坏CD4细胞
掏空了免疫长城 → 严重机会性感染
免疫损伤与并发症

核心问题	大量HIV	免疫缺陷、激活、失衡	OIs与并发症
治疗手段	抗病毒治疗	AAIT	抗感染治疗方案（真菌、TB、细菌等）

AAIT

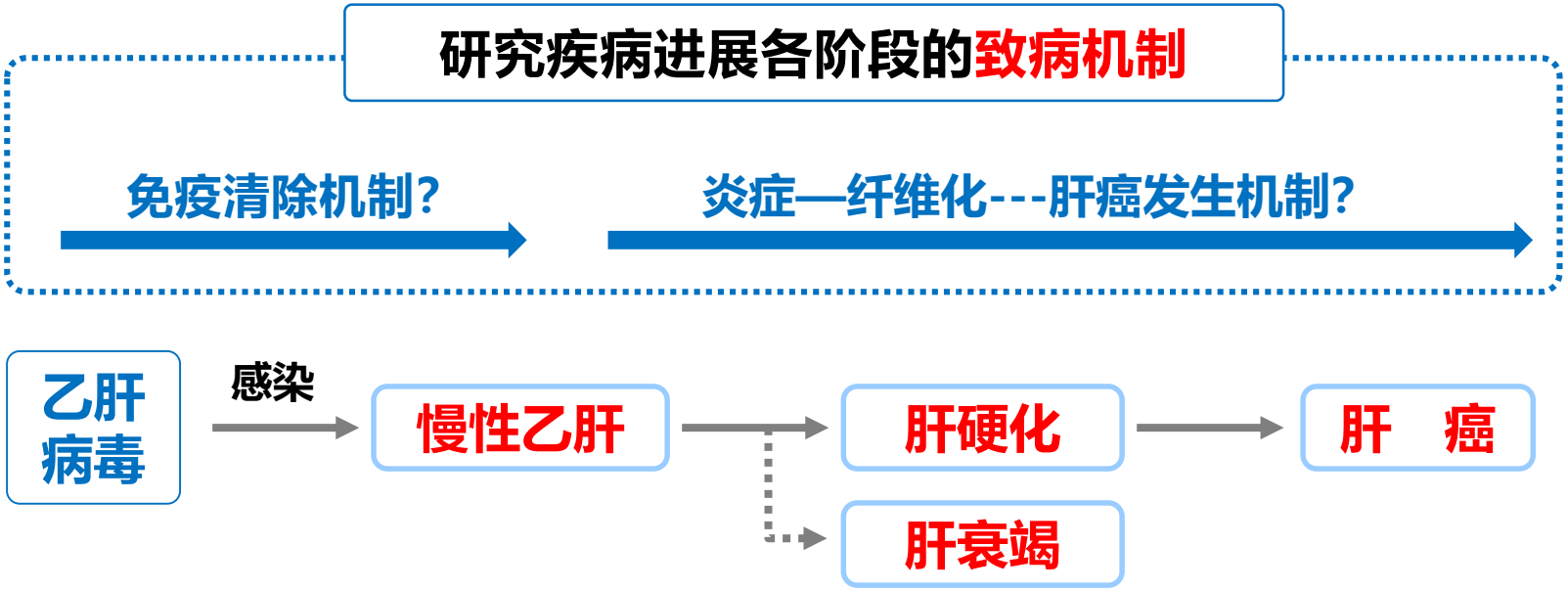
结合临床难题开展转化研究



人体免疫功能

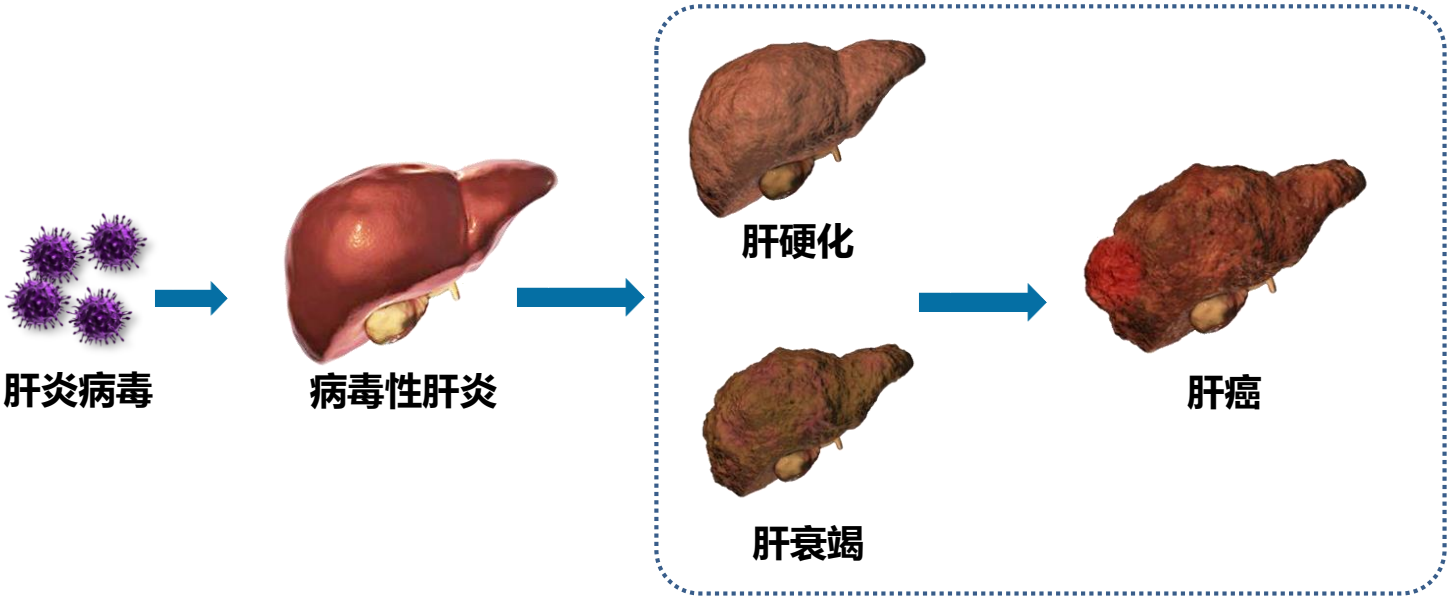
免疫防御	免疫稳定	免疫监视
↓	↓	↓
抗感染免疫	内环境稳定 自体耐受	清除突变、 肿瘤细胞
免疫低下、不能清除HBV	肝细胞坏死 肝组织炎症	肝癌 转移、复发

研究的科学问题（一）



各阶段免疫学特点、抗病毒作用与机制？　？

慢性肝病诊治防面临的挑战



阻断传播

抗病毒治疗/治愈　逆转肝纤维化　阻止终末期肝病

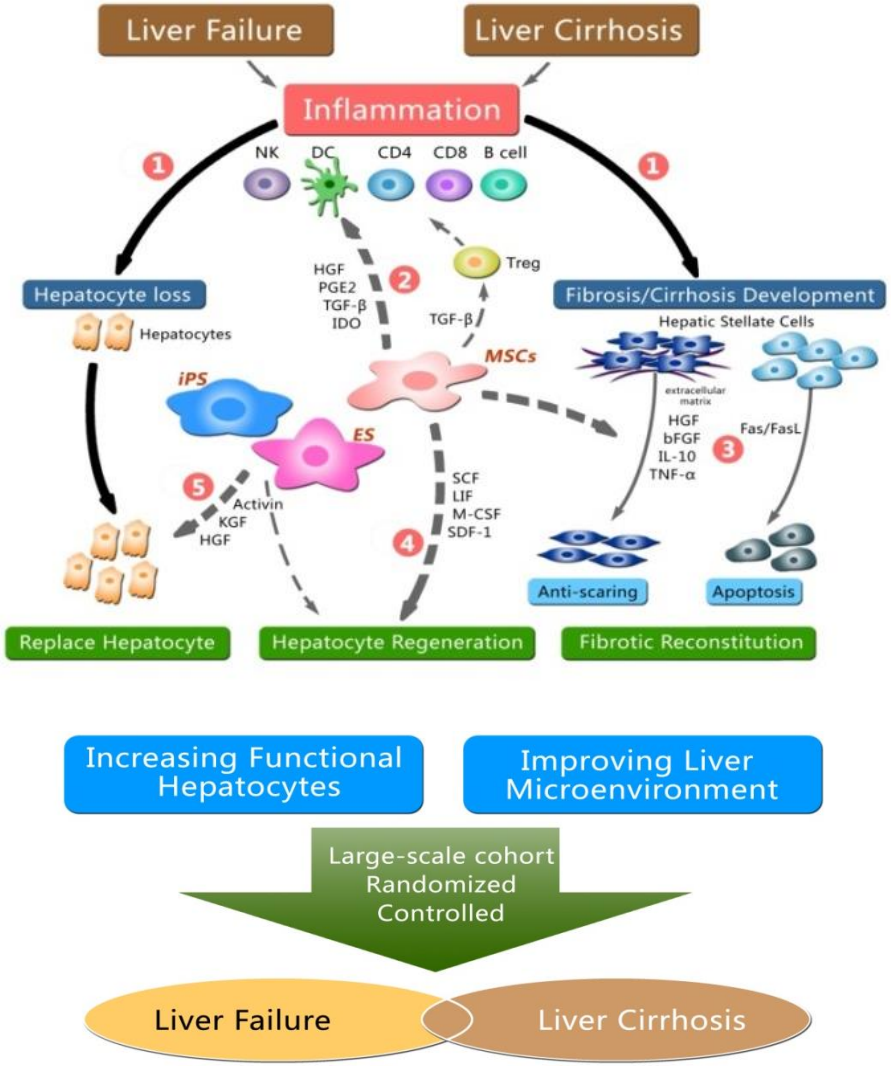
特异性免疫治疗

干细胞治疗

免疫细胞治疗

王福生 等. Hepatol Res, 2007
Exp Rev Gastro Hepatol, 2009

MSC治疗危重肝病临床研究进展与方向



应邀在国际知名杂志撰写专题综述，总结MSC治疗肝硬化和肝衰竭的机制以及今后发展方向。

Zhang Z, Wang FS. J Hepatol , 2013

Li J, et al. Hepatology 2014

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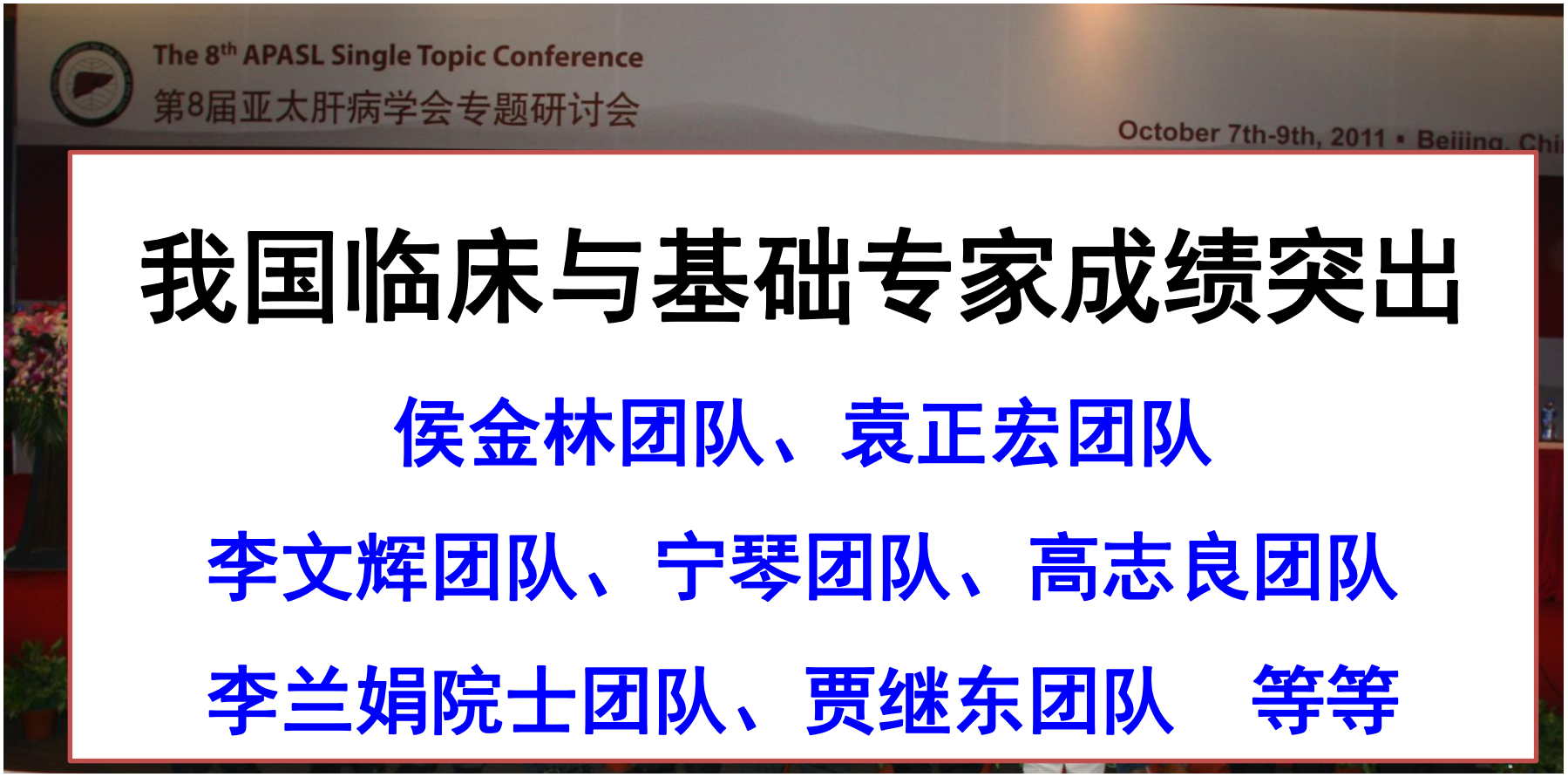
国际合作：作为大会主席举办会议

2011年亚太肝病学会（APASL STC）专题会
肝病临床免疫和免疫治疗大会



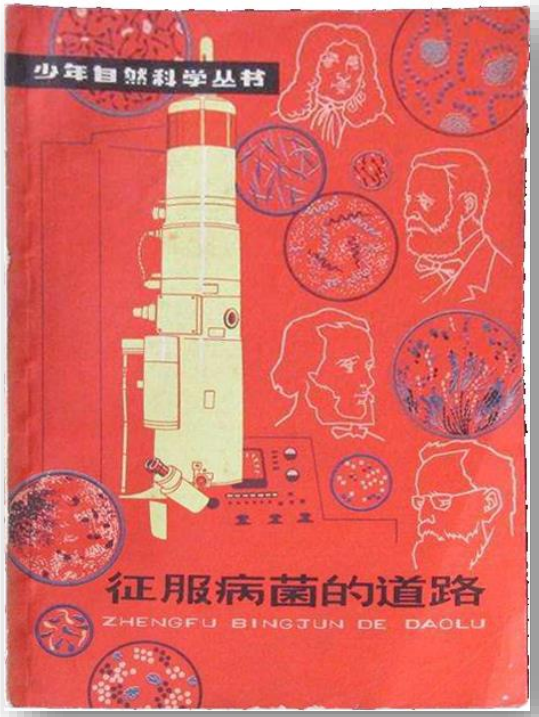
我国临床与基础专家成绩突出

侯金林团队、袁正宏团队

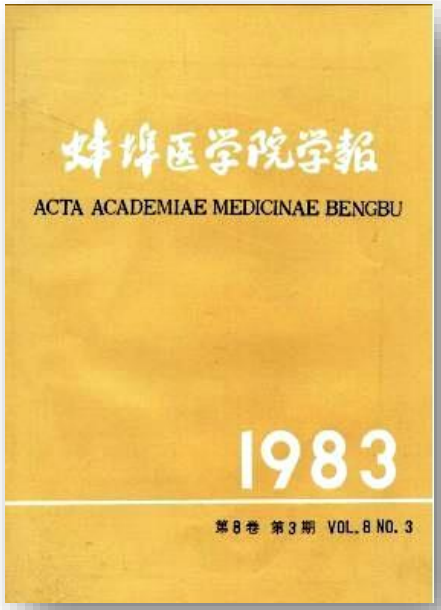
李文辉团队、宁琴团队、高志良团队

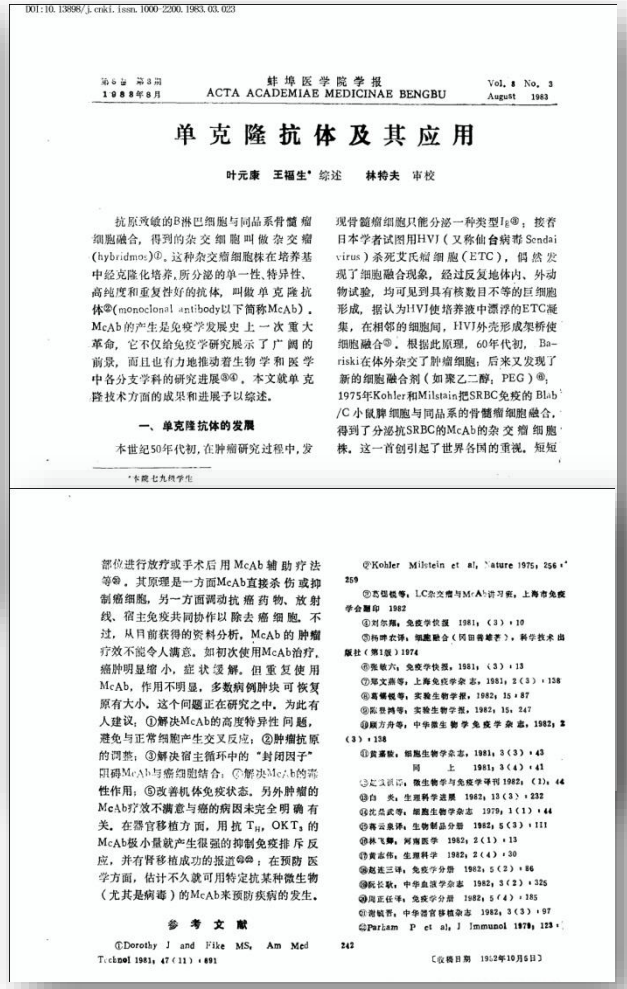
李兰娟院士团队、贾继东团队　等等

不忘初心：对微生物兴趣浓厚



1978年通过阅读科普作品，对微生物和传染病产生浓厚兴趣。




DOI:10.13898/j.cnki.issn.1000-2200.1983.03.023

蚌埠医学院学报 ACTA ACADEMIAE MEDICINAE BENGBU Vol. 8 No. 3 August 1983

单克隆抗体及其应用

叶元康 王福生* 综述 林钧夫 审校

抗原攻敏的B淋巴细胞与同品系骨髓瘤细胞融合，得到的杂交细胞叫做杂交瘤(hybridmoe)①。这种杂交瘤细胞株在培养基中经克隆化培养，所分泌的单一性、特异性、高纯度和重复性好的抗体，叫做单克隆抗体②(monoclonal antibody以下简称McAb)。McAb的产生是免疫学发展史上一次重大革命，它不仅给免疫学研究展示了广阔的前景，而且也有力地推动着生物学和医学中各分支学科的研究进展③④，本文就单克隆技术方面的成果和进展予以综述。

一、单克隆抗体的发展

本世纪50年代初，在肿瘤研究过程中，发现骨髓瘤细胞只能分泌一种类型Ig⑤；按查日本学者试图用HVJ（又称仙台病毒Sendai virus）杀死艾氏瘤细胞（ETC），偶然发现了细胞融合现象，经过反复地体内、外动物试验，均可见到具有核数目不等的巨细胞形成，据认为HVJ使培养液中漂浮的ETC凝集，在相邻的细胞间，HVJ外壳形成架桥使细胞融合⑥。根据此原理，60年代初，Bariski在体外杂交了肿瘤细胞；后来又发现了新的细胞融合剂（如聚乙二醇，PEG）⑦，1975年Kohler和Milstain把SRBC免疫的Blab/C小鼠脾细胞与同品系的骨髓瘤细胞融合，得到了分泌抗SRBC的McAb的杂交瘤细胞株。这一首创引起了世界各国的重视。短短

*本院七九级学生

部位进行放疗或手术后用McAb辅助疗法等⑧，其原理是一方面McAb直接杀伤或抑制癌细胞，另一方面调动抗癌药物、放射线、宿主免疫共同协作以除去癌细胞。不过，从目前获得的资料分析，McAb的肿瘤疗效不能令人满意。如初次使用McAb治疗，癌肿明显缩小，症状缓解，但重复使用McAb，作用不明显，多数病例肿块可恢复原有大小，这个问题正在研究之中。为此有人建议：①解决McAb的高度特异性问题，避免与正常细胞产生交叉反应；②肿瘤抗原的调整；③解决宿主循环中的"封闭因子"阻碍McAb与癌细胞结合；④解决McAb的亲性作用；⑤改善机体免疫状态。另外肿瘤的McAb疗效不满意与癌的病因未完全明确有关。在器官移植方面，用抗T_{11}，OKT_3的McAb极小量就产生很强的抑制免疫排斥反应，并有肾移植成功的报道⑨⑩；在预防医学方面，估计不久就可用特定抗某种微生物（尤其是病毒）的McAb来预防疾病的发生。

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㉑谢毓芳，中华器官移植杂志 1982; 3(3): 97
㉒Parham P et al, J Immunol 1979; 123: 342

〔收稿日期 1982年10月6日〕

1982年，我对单克隆抗体的研究进展产生浓厚兴趣

临床研究：4G-5G时代与时俱进

医疗大数据应用与智慧医疗

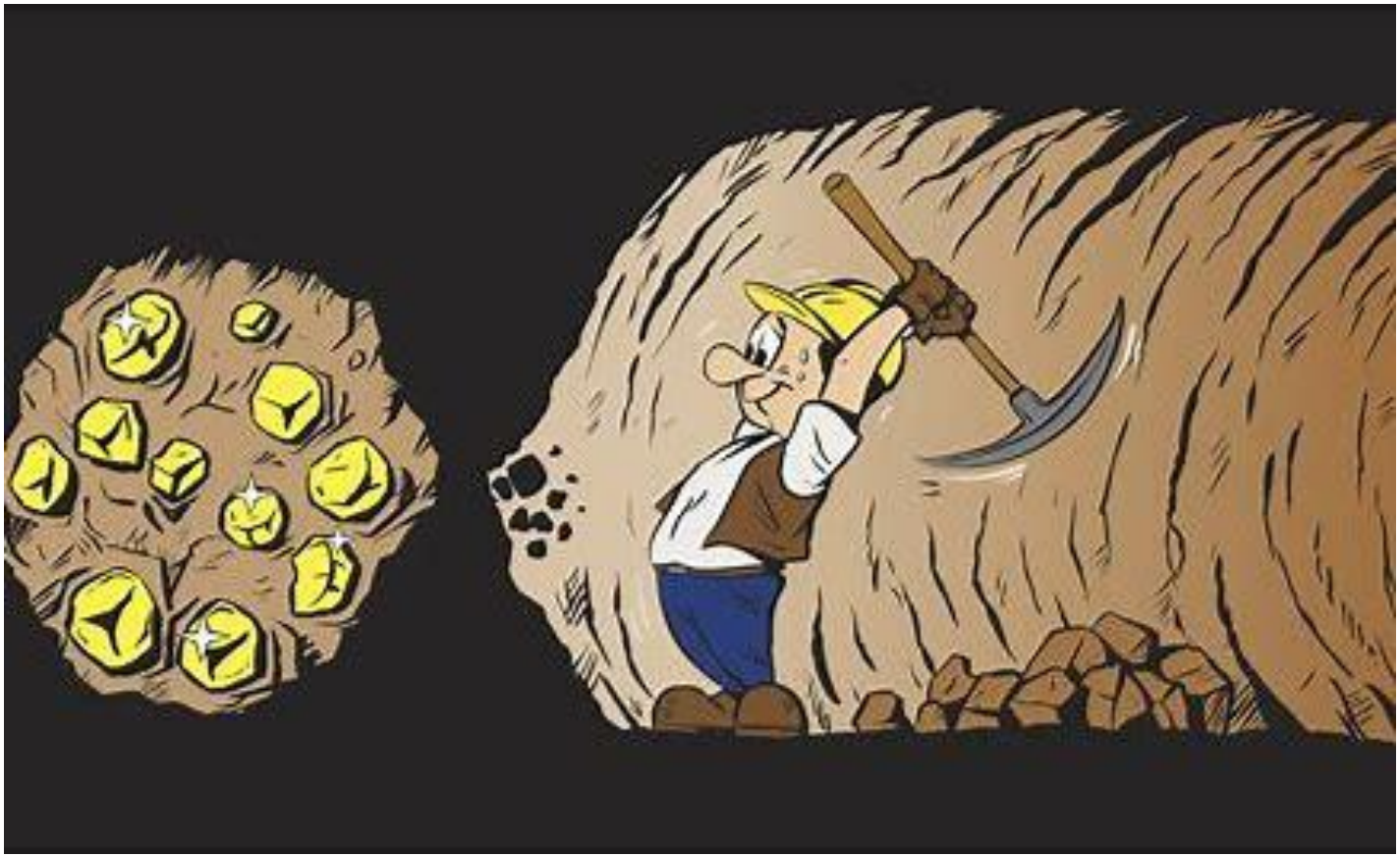
建设—管理—使用

核心问题：人才团队

（医生、护士、辅助科室、统计专家、管理专家等等）

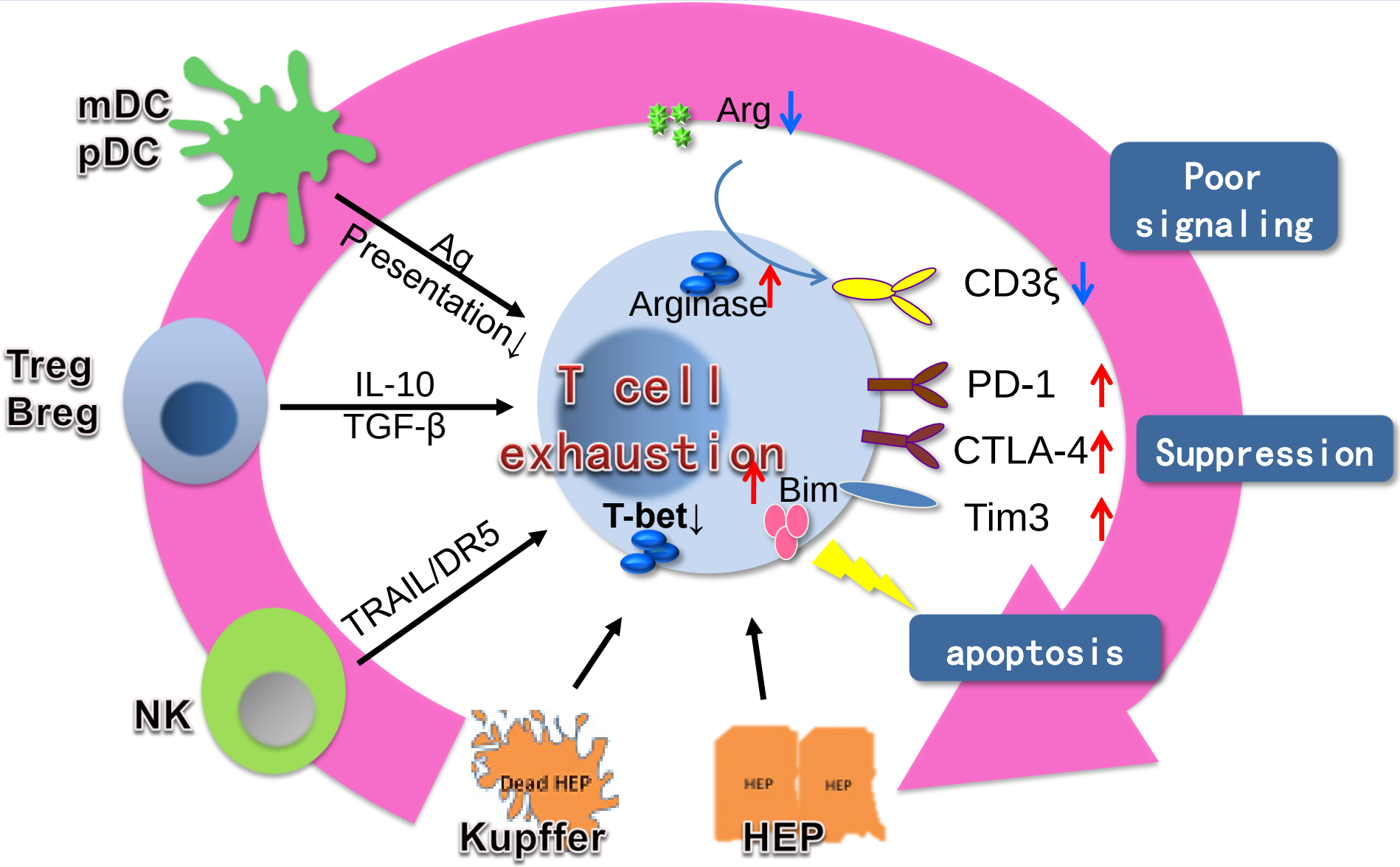
关键问题：全面、系统、完整

如何有效利用好数据：探索本质？



如何破解艾滋病/肝病免疫的八卦阵?

CHB 患者CD8 T细胞功能耗竭



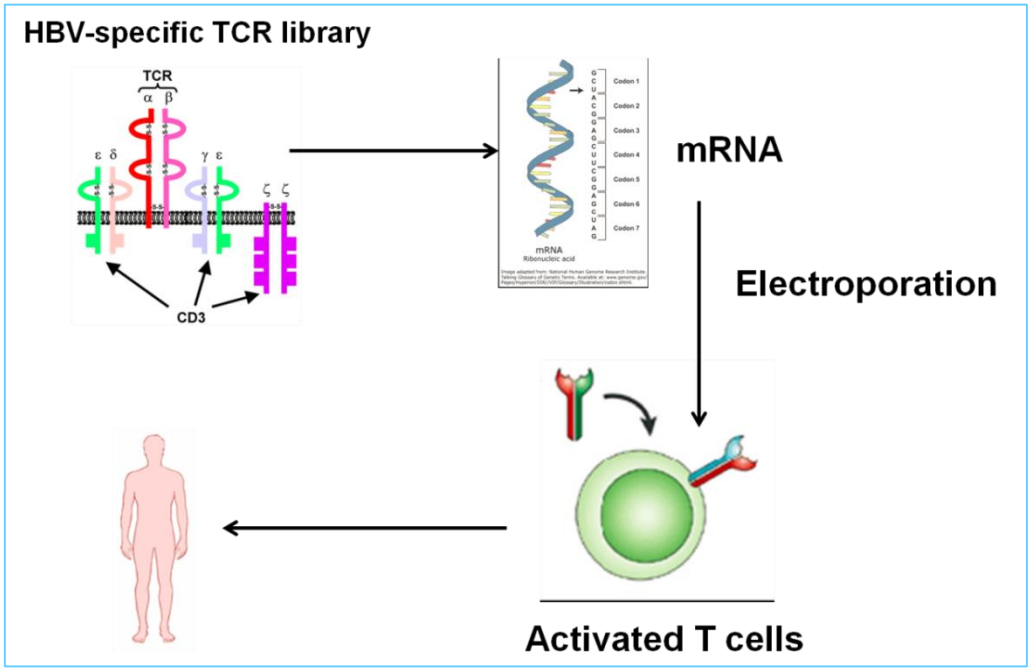
HBV-specific TCR-T cell therapy for HBV

特异免疫治疗有可能克服鸿沟！

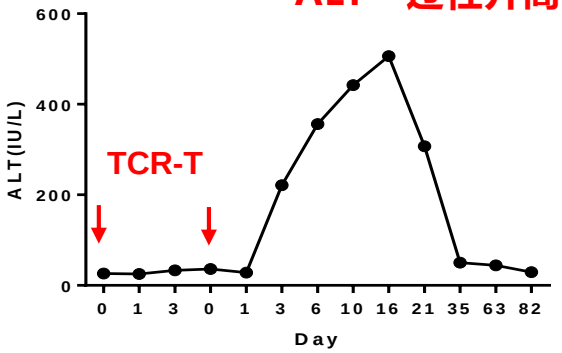
伦理通过、国际注册、知情同意

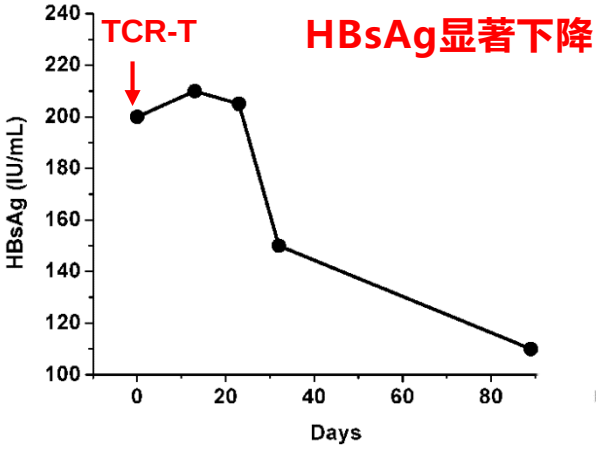
剂量：$1x10^4$/kg　$1x10^5$/kg
时间：2017-2-16　2017-2-23

细胞培养　随访



ALT一过性升高





王福生、施明、Antonio Bertoletti

临床研究数据与感染免疫研究

大样本量、多中心
随机对照临床试验

RCTs 的补充内容（评估 PRO、医疗成本）、行政数据、健康调查、电子健康记录、和医疗图表审查

转化研究

常做的工作

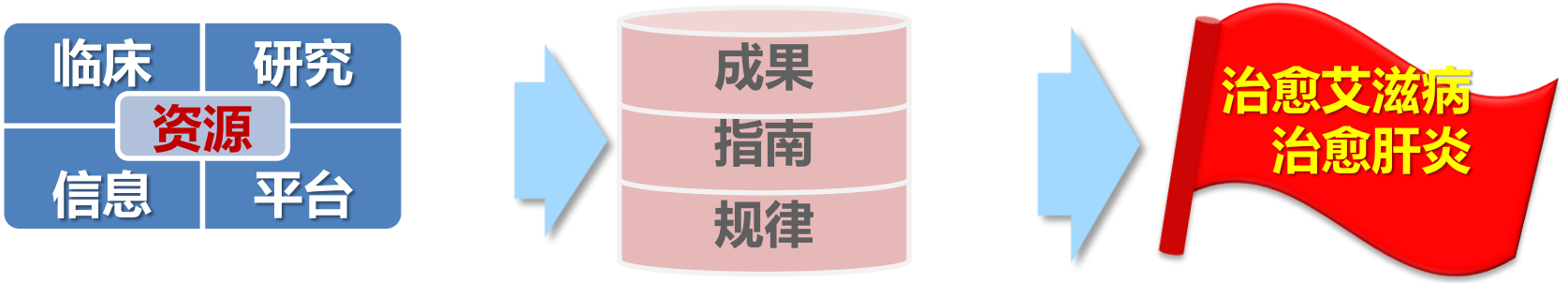
真实世界
临床研究

国家传染病中心建设战略目标

随机对照队列 | **真实世界队列** | **转化研究**

专家团队 | **数据库、样本库** | **方法学平台** | **中心实验室** | **国际合作平台**

出指南、出标准	出技术、出模式	协作平台
规范临床诊疗	创新药物、治疗技术	国内一体化
卫生政策依据	发病机制突破	国际合作化



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Jin-Wen Song
Xing Fan
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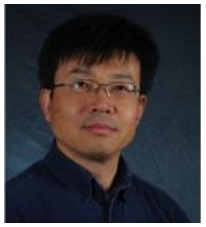

Lishan Su

Yangxin Fu

衷心地感谢各位专家 ！

THANKS!