HCV diagnostics and non-invasive liver disease assessments: what are the current tools in the toolbox and where to from here?

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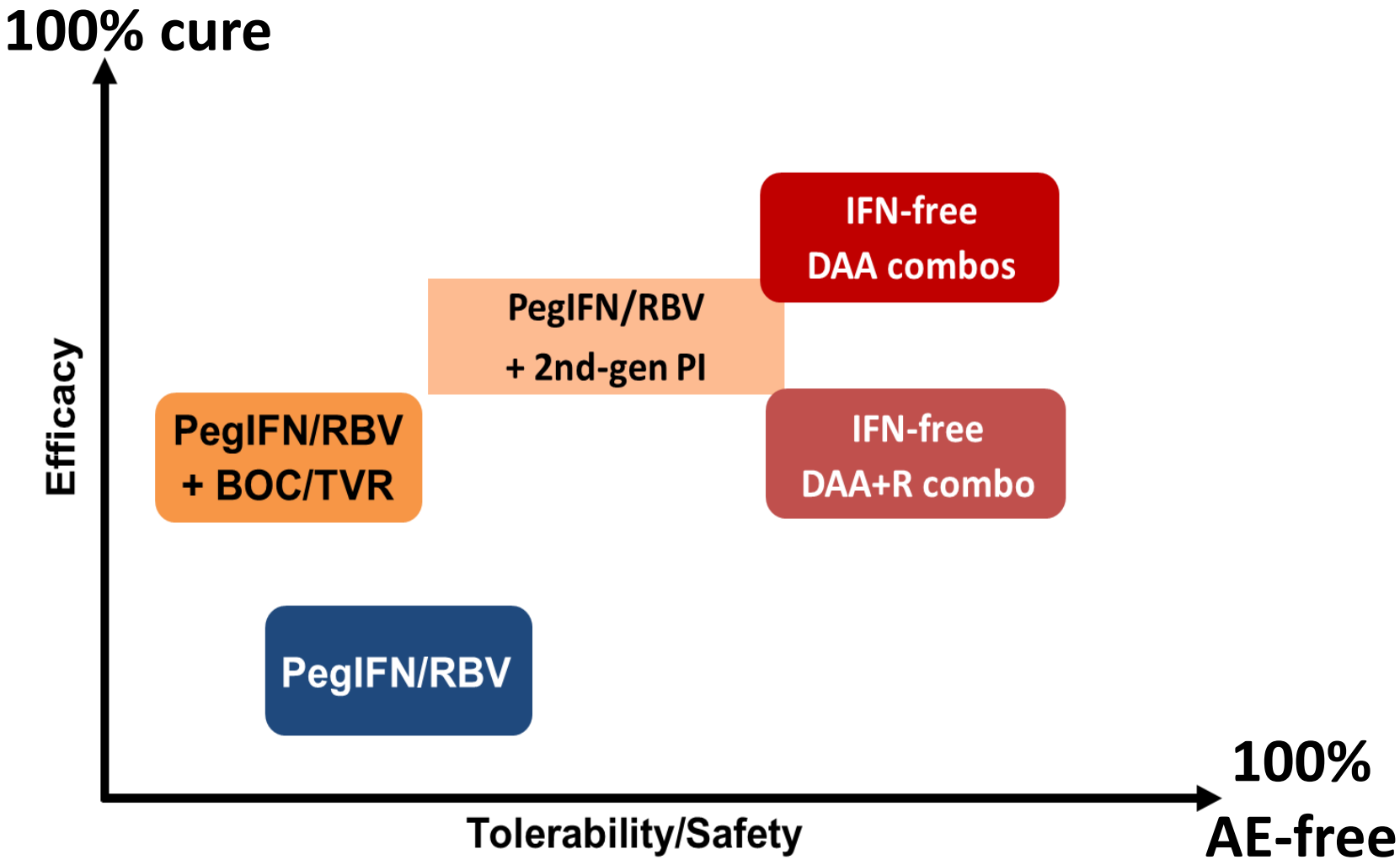
The road towards eradication



Assessment of HCV disease a ide the continuum of

What is the easiest, most cost-effective diagnosis and follow-up strategy for treating the highest number ?

What lies in the tool box?

Tools for the virus (1)

- **HCV Elisa tests[1]:**
  - Standard of care for screening for the presence of HCV Ab
  - Immunoenzymatic assays, 3rd generation
  - Serological window: approx. 60 days
  - May be negative in highly immunosuppressed patients

- **Combo anti-HCV Ab / HCV Ag assay[2]**
  - May reduce the serological window by 20 days
  - Less sensitive than 3rd generation Elisa Tests for Ab and detect HCV later than HCV-RNA in acute HCV settings

[1]Chevaliez S. Clin Microbiol Infect 2011.

Tools for the virus (2)

- **HCV rapid tests**
  - Performed on cravicular liquid, full blood, plasma[1]

Type of tests	Type of liquid	Se [95%CI]	Sp [95%CI]
Oraquick® HCV (Orasure technologies, USA)	Cravicular	97,8% [95,6 - 98,9]	100% [95,6 - 98,9]
	Capillary full blood	99,1% [97,4 - 99,8]	100% [98,0 - 100]
Toyo® HCV (Turklab, Turquie)	Capillary full blood	95,9% [93,1 - 97,8]	98,3% [95,1 - 99,6]
Labmen HCV® (Turklab, Turquie)	Capillary full blood	63,1% [55,1- 70,6]	100% [95,2 - 100]

  - Slight decrease in Se when used on cravicular liquid[2]
  - Controversial effect of HIV on test performance[3,4]

[1]French National Guidelines on HCV Screening, April 2014. [2]Shivkumar, Ann Intern Med 2012. [3]Smith, J Infect Dis 2011. [4]Larrat, J Clin Virol 2012

Tools for the virus (3)

- **HCV-RNA quantification[1]**
  - two molecular biology-based techniques: target amplification (PCR) and signal amplification (branched DNA assay), with an increase in sensitivity in real time PCR
  - Used to confirm chronic hepatitis C
  - In the setting of acute HCV, shortest serological window: 1 – 3 weeks

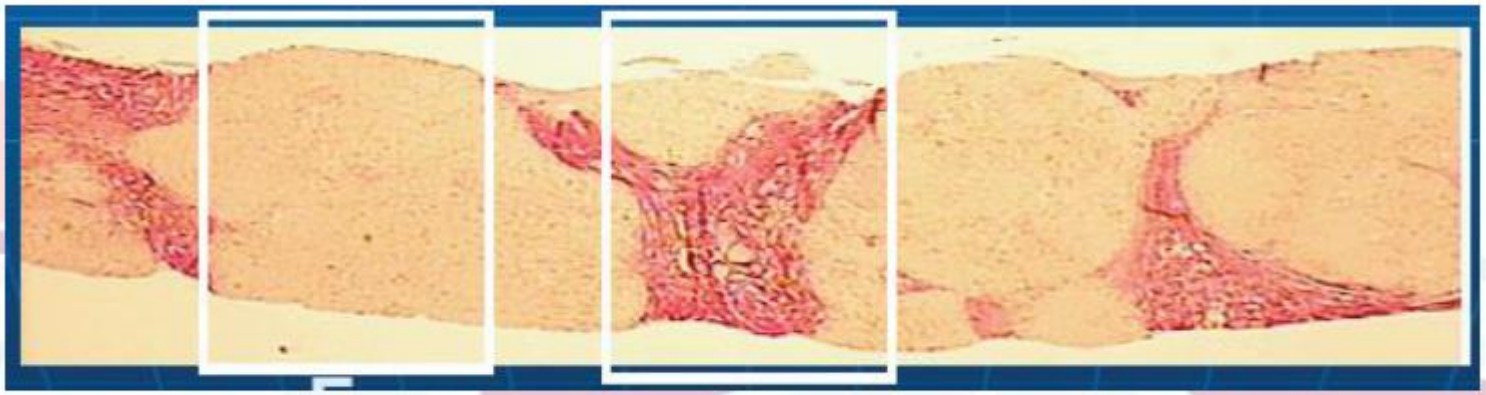
Tools for the virus (4)

- **HCV genotyping**
  - Based on direct sequencing (population sequencing) that provides the full sequence of the analysed fragment, or reverse hybridization that identifies specific nucleotides or motifs at given positions
  - 7 genotypes identified with different susceptibilities to DAAs

[1]Chevaliez S. Clin Microbiol Infect 2011.

Tools for the liver (1)

- **Liver biopsy**
  - Considered as « gold standard » for liver fibrosis evaluation for years
  - Very valuable in HIV patients (NASH, OH, ARV toxicities, etc.)
  - Numerous drawbacks: cost, life-threatening complications, sample variability



(Diapositive empruntée à A. Varaut)

Tools for the liver (2)

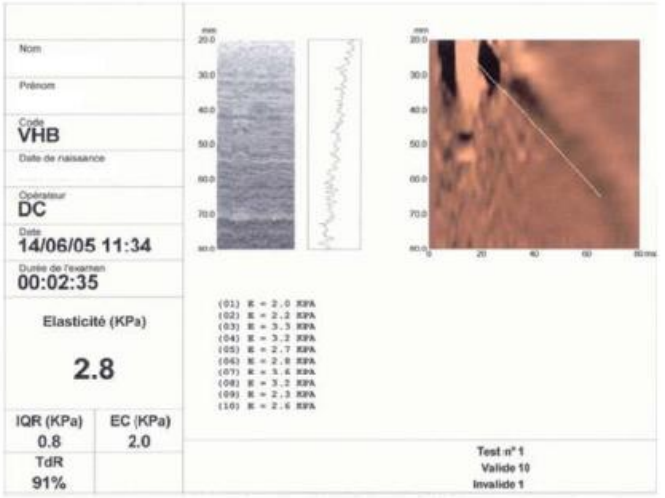
- **Biochemical scores**
  - Based on the combination of biochemical markers with a equation predicting the risk of fibrosis
  - Influenced by etiology of fibrosis (different thresholds)

	Bili	ggt	Hapto	a2M	ApoA	ALAT	Hyalu	Alb	ASAT	Chol	Pqt	TP	Urée	Age	Sexe	IMC
Fibrotest®	x	x	x	x	x									x	x	
SHASTA							x	x	x							
Hepascore	x	x		x			x							x	x	
Zeng		x		x			x							x		
Forns		x								x	x			x		
Fibrometre®				x			x		x		x	x	x	x		
Fib-4						x			x		x			x		
AST/ALT						x			x							
Hyalu							x									
APRI									x		x					
Hui	x							x			x					x

  - Best tests combining markers of extracellular matrix degradation (Fibrotest, fibrometer)
  - Simple tests such as APRI or Fib4 exhibit very good performance in diagnosing advanced fibrosis

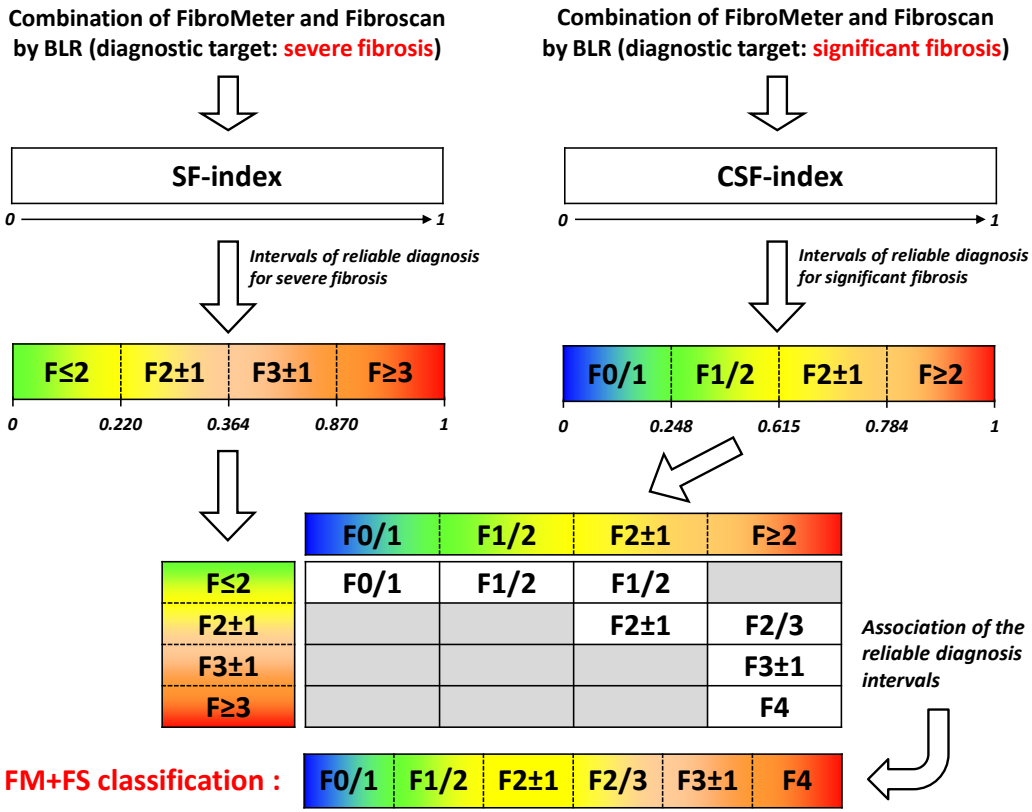
Tools for the liver (3)

- **Transient elastometry**
  - Measurement of liver stiffness (kPa) with a probe
  - Median and IQR with at least 10 valid measures
  - Must be performed after 12 hour-fasting



Tools for the liver (4)

- **Combining non invasive markers for the diagnosis of advanced fibrosis**



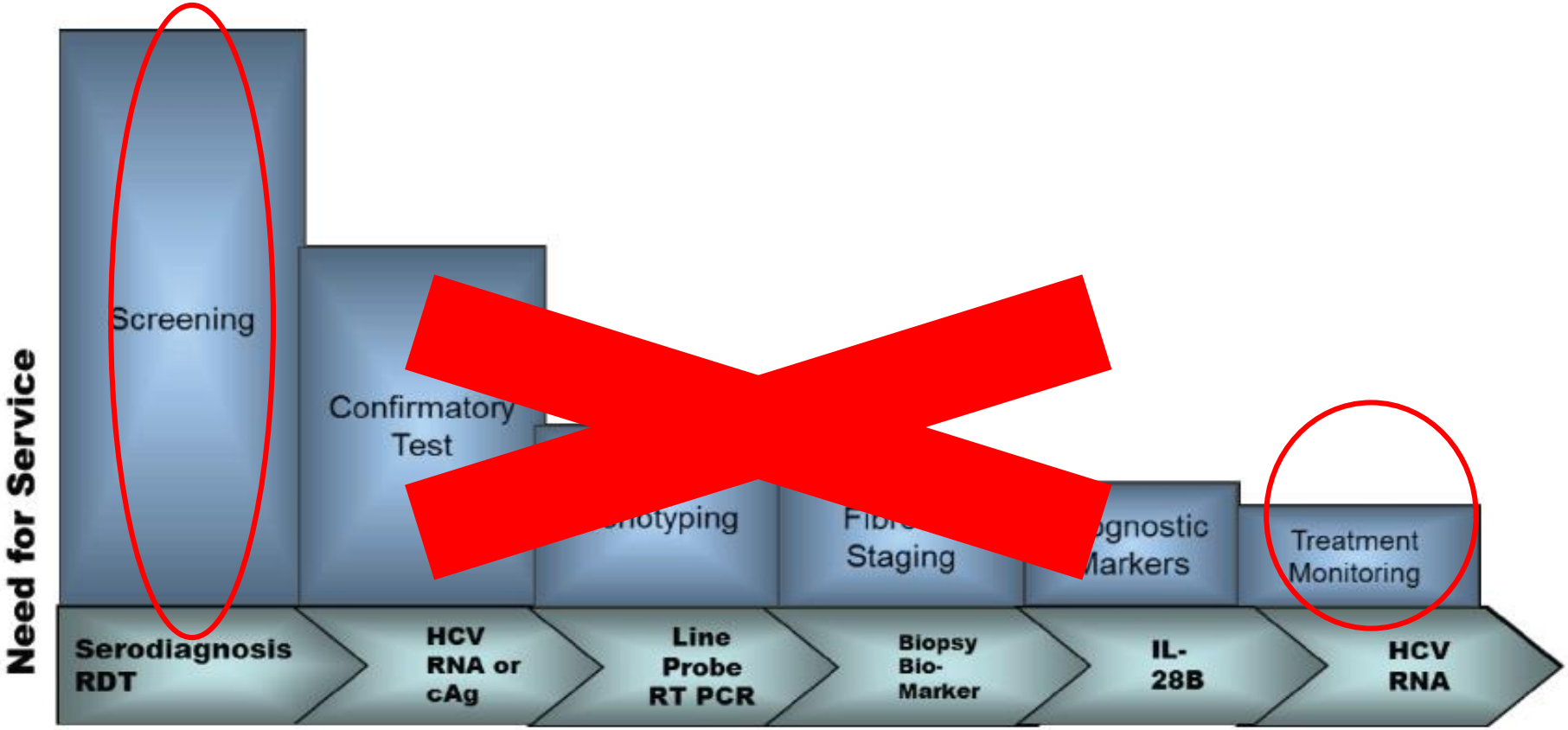
Boursier, Hepatology 2012

Tools for follow-up under treatment

	Before Rx	Rx initiation	W4	W8	EOT	SVR12
FBC	X	X	X	X	X	
LE	X	X	X	X	X	
Renal Fct	X	X	X	X	X	
APRI/TE		X				
HCV-RNA	X	X			X	X
Genotype	X					
Thyroide Fct		X			X	

WHO HCV care and management Guidelines, 2014

How should innovation be a game changer in diagnosis of HCV infection and follow-up of HCV treatment ?



➔ Transforming a multiple-step procedure into a two step-procedure

HCV Core Ag quantification

- **HCV core Ag quantification**
  - Decrease of serological window compared to ELISA, threshold for HCV-RNA detection = 1000UI/mL[1]
  - Excellent Se and Sp which makes it a reliable tool for mass screening for acute HCV[2]
  - Decrease in Se when performed on DBS[3]
  - To date, only one marketed test: Abbott ARCHITECT platform

- But POC device being developed (DAKTARI): eliminates sample preparation through the use of a technology known as “microfluidic immunochromatography”, which isolates cells (or viruses) : the only user step is to apply a drop of whole blood to the cartridge.

[1]Chevaliez, J Clin Virol 2014. [2]Vanhommerig, EASL 2014. Chevaliez, J Infect Dis 2015

ANRS12336: Performance of HCV Core Ag as a screening and follow-up tool

- Ancillary study of TAC trial (efficacy and tolerance of SOF + RBV or SOF + LDV in genotype 1, 2 or 4 HCV infection in Côte d'Ivoire, Cameroon and Senegal)

- Primary objective: performance of HCV Core Ag (Architect Abbott Diagnostics) in HCV screening and follow-up under DAA- based treatment
- Secondary objectives: influence of HBV and HIV coinfection of HCV Core Ag performance

Population

1037 serum samples from the Pasteur Center of Cameroon in Yaounde

Inclusion criteria

- HCV+:
  - HCV antibody (HCV Ab) positive serology
  - Quantifiable HCV RNA
- HCV-:
  - HCV Ab negative serology
  - OR undetectable HCV RNA
- HIV status known
- HBV status known

Exclusion criteria

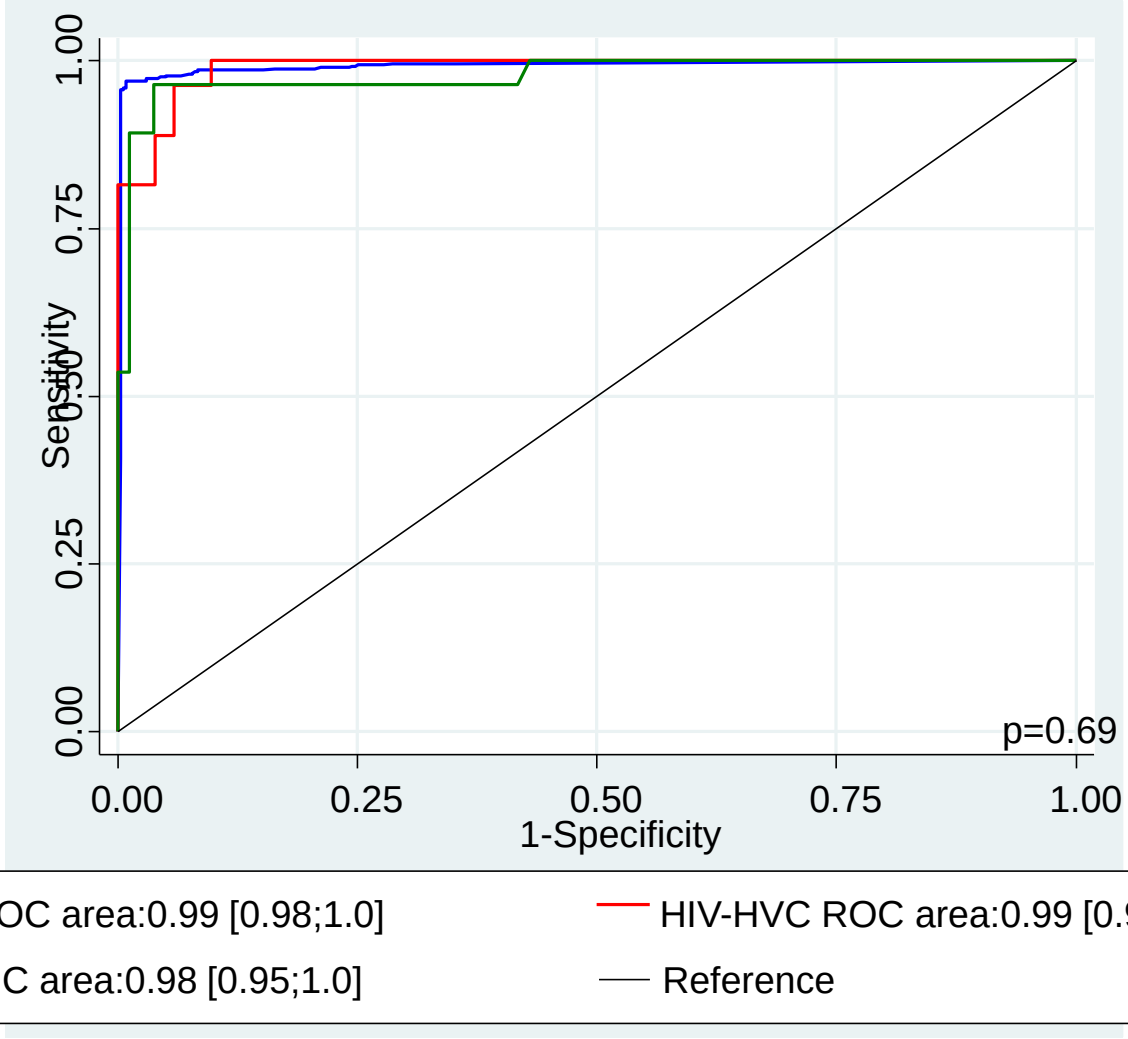
- 11 Tri-infection (HCV/HIV/HBV)
- 7 Infection status unknown
- 10 Retest unavailable

Included samples:
n=1009

- 475 VHC-
- 545 VHC+

Results: AgC overall performance

Figure 2: ROC curves of the performance of AgC quantification for the diagnostic of chronic hepatitis C in HCV mono-infected and HCV uninfected, HIV-infected and HBV-infected patients



Results: overall performance of AgC

Table Performance of the AgC quantification by infection group

	n	Se [IC97.5%]	Spe [IC97.5%]	VPP*	VPN*	AUC [IC95%]	LR+	LR-
Mono	824	95.7 [93.2 ; 97.5]	99.7 [98.1 ; 100]	98.1	99.3	0.99 [0.98-1.0]	319	0.043
HIV	78	100 [85.0 ; 100]	88.2 [74.3 ; 96.2]	57.6	100	0.99 [0.97-1.0]	847	0
HBV	107	96.4 [79.2 ; 99.9]	96.2 [88.1 ; 99.4]	80.2	99.4	0.98 [0.95-1.0]	25	0.037

➔ **Next step: HCV core Ag as a tool for follow-up of patients under treatment**

*Estimated HCV prevalence in Cameroon: 13,8%

HCV RNA quantification

- **Point of care (POC) platforms for HCV-RNA assays**

  HCV RNA quantitative assay

  - Alere Q *(Alere Inc.)*
  - EOSCAPE-HCV rapid RNA assay *(Wave 80 Biosciences)*
  - Truelab Uno real time Micro PCR system *(Molbio Diagnostics Pvt Ltd)*
  - GeneXpert *(Cepheid)*
  - RT CPA HCV Viral Load Test *(Ustar B*

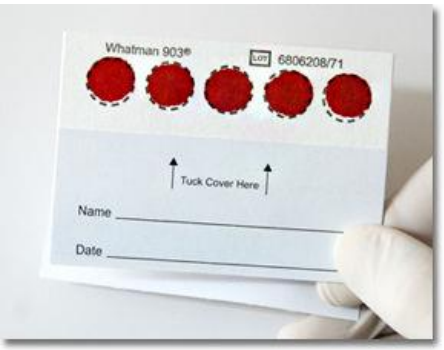
  HCV RNA qualitative assays

  - Gendrive *(Epistem)*
  - PanNAT *(Micronics Inc.)*

Source: UNITAID report April 2015

HCV Genotyping

- POC molecular devices under development, i.e Gendrive (EPISTEM)



Dried Blood Spots

- Used to collect venous blood specimens in setting where syringes, tubes, centrifuges and skilled labor are not available
- Recently assessed for a wide range of HCV diagnostic tools[1]:

Variable	Specificity, % (95% CI)	Sensitivity, % (95% CI)	PPV	NPV
Anti-HCV antibody detection	98.2 (94.9–99.6)	99.1 (97.4–99.8)	99.1	98.2
HCV core antigen detection	100 (97.8–100)	64.1 (58.5–69.3)	100	64.7
HCV RNA detection				
CAP/CTM	100 (97.8–100)	97.1 (94.7–98.5)	100	95.0
*m*2000	100 (97.8–100)	98.1 (95.9–99.1)	100	96.6
HCV genotype determination	NA	72.3 (67.0–76.9)	100	NA

Results of serum analysis are the references.

Abbreviations: CAP/CTM: Cobas Ampliprep/Cobas TaqMan HCV assay, version 2; CI, confidence interval; *m*2000, *m*2000 platform; NA, not applicable; NPV, negative predictive value; PPV, positive predictive value.

Soulier, J Infect Dis, 2015

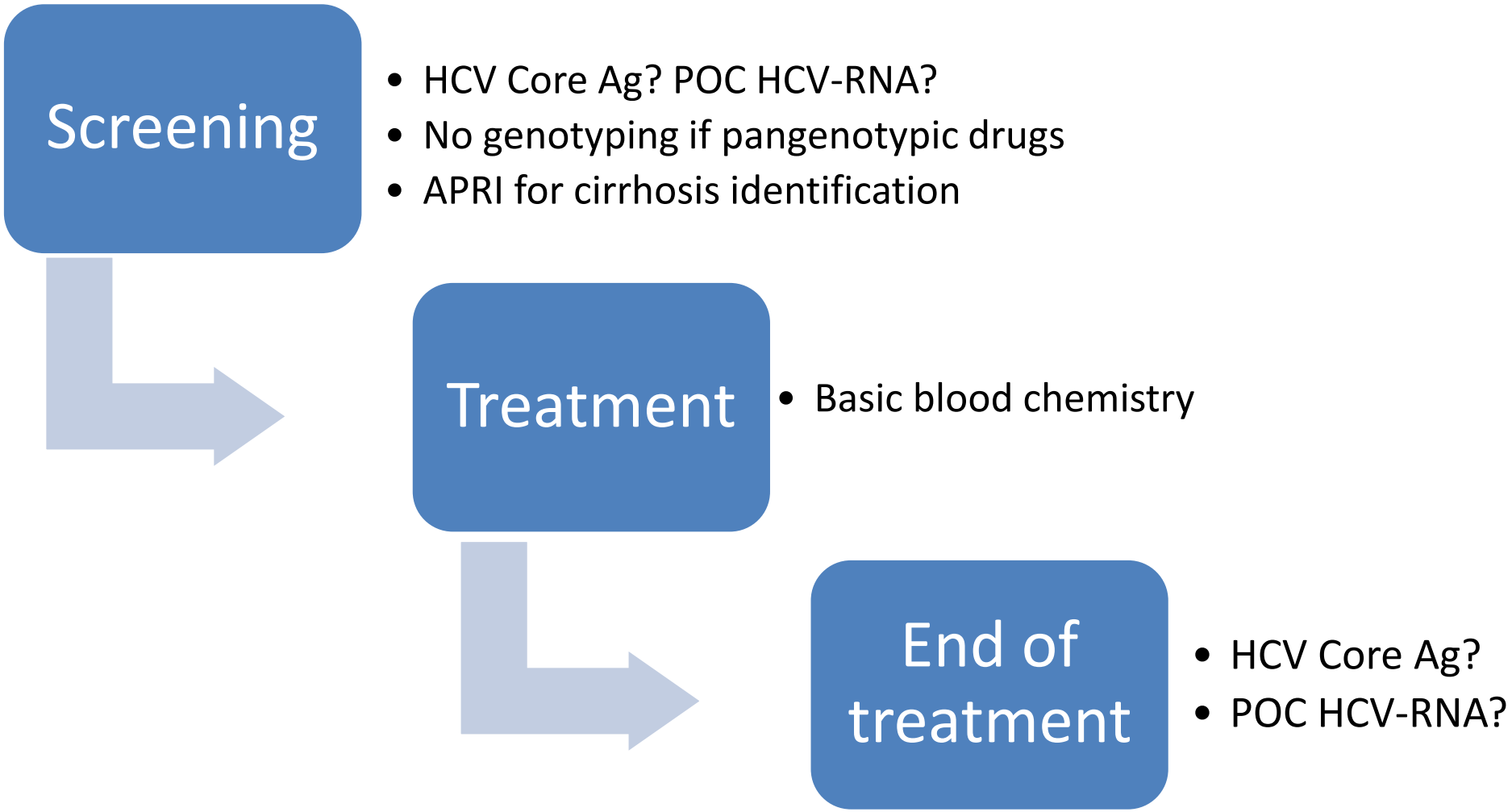
The use of APRI for identifying cirrhosis

		APRI (low cut-off)	APRI (high cut-off)	FIB4 (low cut-off)	FIB4 (high cut-off)	Transient elastography (Fibroscan)
Significant fibrosis (METAVIR ≥F2)	Sensitivity (95% CI)	82 (77–86)	39 (32–47)	89 (79–95)	59 (43–73)	79 (74–84)
	Specificity (95% CI)	57 (49–65)	92 (89–94)	42 (25–61)	74 (56–87)	83 (77–88)
Cirrhosis (METAVIR F4)	Sensitivity (95% CI)	77 (73–81)	48 (41–56)	–	–	89 (84-92)
	Specificity (95% CI)	78 (74–81)	94 (91–95)	–	–	91 (89–93)

APRI aminotransferase/platelet ratio index; kPa kilopascal

Source: WHO HCV Guidelines, April 2014

What would the ideal screening and follow-up algorithm?



Evaluation of a simplified screening and follow-up strategy in a real-life setting

- Nested in two studies based in Western Africa:
  - TAC (Treatment Africa Hepatitis C) ANRS12311
    - Clinical trial assessing the efficacy and tolerance of a 12 week-course of SOF+RBV or SOF+LDV in 120 GT1,2 or 4 patients living in Cameroon, Côte d'Ivoire and Senegal
      - Of whom 20 are DUs living in Dakar on OST provided at CEPIAD[1]
  - CODISEN (Cohort of Drug Injectors living in Senegal) ANRS12334
    - Cohort of 500 individuals seeking care at the CEPIAD OST center (HCV prevalence: 23,3%, 38,3% in injectors)
    - Access to TAC trial and MRKHEPSEN (Gazoprevir+ elbasvir for 12weeks in GT1 and 4)

[1]Lepretre, JIAS 2015

Acknowlegments

- TAC clinical research team and patients
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