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Reflection paper on regulatory requirements for the development of medicinal products for non-alcoholic steatohepatitis (NASH)

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1. Introduction

Non-alcoholic steatohepatitis (NASH) is a disease of unmet medical needs. At the same time, the specifics of the diseases create major challenges for the development of new medicinal products. This reflection paper describes the current regulatory approach in the EU with respect to NASH.

Problems raised and potential solutions described in this reflection paper, may only partly be transferable to other chronic liver diseases. Potential applicants are advised to seek scientific advice before translating the statements of this paper to other chronic liver diseases.

2. Scope

As a reflection paper, this document provides a high-level description of the requirements for drug development in the field. For NASH, the regulatory experience with the licensing of new medicinal products is limited. Therefore, this paper aims at a preliminary definition of development strategies, which, in the case of successful marketing authorization applications occurring in the future, will have to be refined, and may finally be superseded by a full guidance document. Due to the unmet medical need, and increasing health burden of the disease, the development of therapies addressing this unmet need is of public relevance.

This paper concentrates on developments for a standard, biopsy-diagnosed patient population (see Chapter 4 and 5.2.2). In clinical practice, there is a certain degree of disconnect between the strict definition of NASH based on histology criteria and routine diagnosis (especially in non-hepatology practice). For the time being, potential developments in completely non-invasively diagnosed patient populations (including the conduct of outcome trials) are not within the scope of this reflection paper. Applicants intending such a development are recommended to apply for Scientific Advice.

3. Legal basis and relevant guidelines

This document should be read in conjunction with the introduction and general principles and part I and II of the Annex I to Directive 2001/83/EC as amended. Applicants should also refer to other relevant EU and ICH guidelines (in their current version) and regulations, especially the following:

- Reflection paper on assessment of cardiovascular safety profile of medicinal products (EMA/CHMP/50549/2015)
- Reflection paper on the use of extrapolation in the development of medicines for paediatrics. (EMA/189724/2018)
- Guideline on clinical development of fixed combination medicinal products. (EMA/CHMP/158268/2017)
- Points to consider on application with 1. Meta-Analyses; 2. One pivotal study. CPMP/EWP/2330/99.
- ICH E9(R1) Addendum on estimands and Sensitivity Analysis in Clinical Trials (EMA/CHMP/ICH/436221/2017)
- Guideline on the evaluation of pharmacokinetics of medicinal products in patients with impaired hepatic function CPMP/EWP/2339/02
- Guideline on clinical evaluation of medicinal products used in weight management EMA/CHMP/311805/2014

4. Background on non-alcoholic steatohepatitis

NASH is considered the progressive, necro-inflammatory phenotype of non-alcoholic fatty liver disease (NAFLD)[1], which itself is the most prevalent chronic liver disease worldwide with an estimated prevalence in the Western world of around 25%[23], and it is estimated that about 20-50% of these suffer from NASH[45]. The progression from ("simple") fatty liver to a progressive form of disease is thought to be related to the development of liver cell stress, subsequent inflammation, and fibrosis with the potential development of cirrhosis and end-stage liver disease (ESLD) with increased risk of hepatocellular carcinoma (HCC). NASH associated ESLD is expected to represent the highest share of patients referred for liver transplantation in the USA in the future[6] and the disease burden and economic impact are expected to reach similar levels in Europe[789].

NASH is associated with other comorbidities and (metabolic) risk factors such as obesity, arterial hypertension, diabetes mellitus type 2 (T2DM), atherogenic dyslipidaemia, and others. The disease – although genetic factors have also been identified and the pathophysiology is complex and still incompletely understood [101112] – is thought to be largely the consequence of hyperalimentation and so-called Western diet and has been regarded to be the hepatic manifestation of the so-called metabolic syndrome and is regarded to be a world-wide problem[1314].

A proposal for the re-labelling of NAFLD as "Metabolic Associated Fatty Liver Disease" (MAFLD) has been made and is related to the close relationship to over-alimentation and metabolic dysfunction but also to the avoidance of potentially stigmatising nomenclature[1516]. A multi-society Delphi consensus has finally concluded, that NAFLD should be renamed as "metabolic dysfunction-associated steatotic liver disease" (MASLD), as well as to relabel NASH as "metabolic dysfunction-associated steatohepatitis" (MASH). The main aspect of this relabelling refers to MASLD, which was redefined and will compulsorily require the presence of 1 out of 5 cardiometabolic risk factors. The term MASH, contrary to MASLD does not include a revision of the definition and still includes the term steatohepatitis and is intended to ensure retention and validity of prior data from clinical studies [17].

Therefore, while "positive" diagnostic criteria are clearly applicable for MASLD and this is no longer a diagnosis of exclusion only, the diagnosis of NASH will mainly remain a diagnosis of exclusion (notably, infectious and non-infectious other liver disease) requiring confirmation by liver biopsy, referring to the histologic features steatosis, hepatocellular ballooning, lobular inflammation, and fibrosis[18].

In the following, the term NASH will be used, but it is expected that this will consecutively be replaced by the term MASH by all stakeholders in the future.

Although health-related quality of life may be impaired[19], symptoms do not play a clinically important role in the diagnosis of (non-cirrhotic) NASH and there are no symptoms thought to be specific for the disease. Also the awareness with regard to the disease and of the associated risks is poor[20].

The natural history of NASH has not been fully elucidated, and further efforts are needed to clarify important aspects, e.g., overlap of progression and regression[21]. The risk of progression to ESLD is largely related to the baseline fibrosis grade[22]. The progression of fibrosis is estimated to be a slow process taking years for one fibrosis stage, and the development of ESLD. [232425262728].

5. Recommendations

5.1. General considerations on regulatory strategy

Based on its slow progression without prominent and specific symptoms, NASH is difficult to study for long-term outcomes over a reasonable time span. The term "long-term outcome" is used in the

following for events such as liver transplantation and death, as well as clinical events of decompensation of liver cirrhosis, which are otherwise also termed "hard outcomes".

An acceptable regulatory strategy for applicants developing new agents in the disease area may be to define intermediate endpoints for which a reasonable assumption for the prediction of long-term outcomes can be made. These reasonable assumptions are usually based on associations with risk factors for the long-term outcomes in observational natural history cohorts and the biological plausibility attributed. The term "intermediate endpoint" will be used throughout in the following for events otherwise also termed "interim" endpoints.

Strictly speaking, however, such endpoints are not validated in the sense that positive changes for the intermediate as well as the long-term clinical outcome have repeatedly and consistently been demonstrated for therapeutics. Due to the unmet medical need in the field, a strategy to obtain an early, conditional approval (=conditional marketing authorisation; CMA) of new compounds based on these intermediate endpoints could be considered. This strategy will require the confirmation of efficacy (and safety) of the compound after approval, documenting the effects on long-term outcomes. Such a strategy, however, is only acceptable if an unmet need is still present[29], a positive benefit-risk ratio can be concluded, and it is likely that the applicant will be in a position to provide comprehensive clinical data post-marketing.[30] The potential obstacles for continuation of confirmatory studies after early approval will, however, need to be considered (see Chapters 5.3.1 and 5.3.2).

The acceptance of the mentioned regulatory strategy for CMA [31] will be evaluated on a case-by-case basis.

This reflection paper outlines currently acceptable intermediate (for the manifestation of ESLD), as well as suitable long-term endpoints.

Acceptable intermediate endpoints are currently mainly based on the histological evaluation of liver biopsies. Liver biopsy and histology have been widely criticized for sampling error and intra- and inter-observer variability[32]. However, potential non-invasive methods proposed to replace histology as intermediate endpoints, are still insufficiently validated for NASH. Therefore, histology is in most cases still the state of the art for the diagnosis of non-cirrhotic NASH and compensated cirrhotic NASH as well as for the follow-up of the course of the disease, for the purpose of clinical studies.

As liver biopsy comes with a significant patient burden, invasiveness, and the associated risks of morbidity[33] and potentially even mortality, this reflection paper calls for the further development of non-invasive methods to replace liver histology in the future. Serological markers or imaging methods can be regarded as promising candidates. Drug developers are therefore encouraged to aim at producing evidence for future validation of novel methodologies intended to replace histology in the future within their drug development programs. For the "validation" of non-invasive intermediates, it may be necessary to generate data with long-term observations, including the occurrence of clinical outcomes, with liver decompensation events, transplantation, and death[34].

Possible targets of estimation that define treatment effects of interest in NASH (according to the ICH E9 R1 addendum will also be described in this reflection paper.

5.2. Selection of patient populations

The usual principles of the selection of study population, such as being representative of the target population in terms of demographic characteristics and co-morbidities are of course applicable to NASH. The diagnosis of NASH is a diagnosis of mixed exclusion of other relevant diseases, as well as a positive diagnosis, which is mainly reliant on liver biopsy with histology.

5.2.1. Clinical characteristics

A selection of patients on the basis of symptoms is usually not possible. With the new nomenclature it is expected that all MASH/NASH patients will also be diagnosed with MASLD and therefore have at least 1 out of 5 cardiometabolic risk factors (obesity, diabetes/insulin resistance, arterial hypertension, hypertriglyceridaemia, hypercholesterolaemia).

Since most patients with NASH will be obese, and the positive influence of weight reduction on NASH has clearly been demonstrated, weight and weight reduction are important variables for the further course of the disease. At the time of inclusion (=randomisation), patients should, however, rather be requested to have stable weight for a certain timeframe.

Co-morbidities, such as diabetes, dyslipidaemia, and hypertension should be treated adequately and stably at the time of inclusion. Since medication for the treatment of type 2 diabetes mellitus and obesity have a potential for influencing the disease process in NASH, considerations on in- or exclusion, timing, and dosing of such medication may be relevant, especially in case of the need for additional treatment during the study. Clear instructions for handling of the treatment of co-morbidities need to be defined in the protocol. The best acceptable strategy is to investigate an investigational treatment on top of standard of care. Stratification for such factors is advisable to allow a balanced evaluation of these covariates.

Other chronic liver disease (such as viral hepatitis, PBC, PSC, autoimmune hepatitis, Wilson's disease, etc.) should be excluded. In addition, the exclusion of relevant alcohol intake is, according to the definition of the disease, required for the inclusion of patients into clinical studies. This should be based on validated questionnaires, lifetime history of alcohol intake, as well as relevant biomarkers. The acceptable alcohol intake has been defined as 20 g or 2 units/day in women, 30 g or 3 units/day in men and is usually defined as low or light alcohol intake[35]. Adequate cut-offs for the biomarker-based evaluation of alcohol intake, depending on the biomarker chosen, should also be defined. Monitoring of alcohol intake (e.g., biomarker based) is recommended during clinical studies.

5.2.2. Biopsies and histology

Histology is currently considered the gold standard for finally securing the diagnosis, as well as determining the severity of disease, and is also recommended as part of clinical practice. There is currently a broad consensus (e.g., see the multi-stakeholder composed Liver Forum publications[36]) that histology should always be available, also in early clinical studies, and inclusion of patients should generally be based on histological evaluation (grading and staging). Deviations for exploratory clinical studies, e.g., using imaging methods, or biomarkers (or a combination of those) only, are possible if based on sound scientific principles, for which the uncertainties can be quantified, and later stage studies be planned accordingly. For the cirrhotic population, see Chapter 5.2.4.

The indication wording is determined at the time of marketing authorisation application (MAA) after assessment of the full data. However, since the mainstay of data will conceivably be generated in a population diagnosed with biopsy, this may have influence on the final wording of the indication. A mix of biopsy- and non-biopsy based patient populations and consistency of results across these groups may be needed to allow an unrestricted indication wording.

Biopsies should generally not be older than 6 months at the time of inclusion into a clinical study (for potential exceptions, see below). The risk of progression to ESLD, liver transplantation and death has been demonstrated to be independently associated with the stage of liver fibrosis, with only minimally increased risk for stage 1 patients[37]. Fibrosis stage 1 patients are therefore currently only

recommended for inclusion in therapeutic studies in NASH for exploratory purposes. The study population is therefore expected to include patients with fibrosis stages 2-4.

For the histological diagnosis for inclusion (and evaluation of histology endpoints) the NASH-clinical research network (CRN) grading system is the recommended grading system[38]. However, patients may also be included (and evaluated) based on potentially other grading systems for NASH (e.g. SAF score[39]), provided the validation of respective grading systems is substantiated.

The inter- and intra-observer variability for some of the features of the CRN-system has been revealed to be relevant[40], and efforts to overcome the weaknesses of the scoring system[41], including the use of artificial intelligence-aided methods are in principle welcomed from a regulatory perspective. However, since alternative methods have not been fully validated yet, applicants wishing to use different methodologies are advised to seek scientific advice. For the "conventional" evaluation of histology, in order to limit variability of the methods, use of centralised evaluation by at least two experienced histopathologists, including algorithms for arbitration is recommended.

In the following, specific criteria for inclusion into studies in NASH will be dealt with dividing the populations into those with or without the presence of liver cirrhosis.

5.2.3. Non-cirrhotic NASH (fibrosis stage 2 and 3)

The selection of patients in fibrosis stages 2 and 3 studies should be based on the full evaluation of histology including the feature for disease activity and grading because developments of regression and progression may overlap, and the risk of progression has also been associated with higher degrees of ballooning and inflammation. If the NAS-CRN system is used, a total NAS of greater or equal than 4 with at least a score of 1 for ballooning and lobular inflammation each should be used.

Currently, inclusion of patients based on non-invasive methods excluding biopsy/histology based on clinical features, imaging, and/or biomarkers in confirmatory studies is generally not recommended[42]. However, the use of clinical features and non-invasive methods to identify a high proportion of patients fulfilling the histology criteria during screening are recommended to avoid unnecessary biopsies.

5.2.4. Compensated cirrhotic NASH (fibrosis stage 4)

In patients with manifest cirrhosis (=fibrosis stage 4), the presence of a rigorous minimal grade (for steatosis, inflammation and ballooning) is less critical, because the risk of (clinical) progression is considered to be high, based on the presence of cirrhosis alone. Nevertheless, these features may still be present and can be used as inclusion criteria in similar way as for the non-cirrhotic population. However, due to the ongoing remodelling process of the liver, these features might get "lost" over time.

In these so-called "burnt-out NASH cirrhosis" or patients initially diagnosed with cryptogenic cirrhosis[43], in case definite NASH is not present, all of the following features should be documented in order to make the diagnosis NASH sufficiently likely: Historical biopsies with presence of unequivocal NASH, a high likelihood of NASH based on non-invasive testing (biomarker and imaging; criteria need to be clearly defined), and the presence of associated co-morbidity (e.g. at least two co-morbidities such as obesity, type 2 diabetes mellitus, or hyperlipidaemia). A "qualitative" scoring for the likelihood of the presence of NASH-associated cirrhosis has been presented[44]. However, since the quoted likelihood has not been quantified, applicants are advised to seek advice in case less stringent criteria are intended to be used.

While for these patients, a biopsy demonstrating cirrhosis (fibrosis stage 4) is usually required, cirrhosis is usually diagnosed non-invasively in clinical routine. The criteria for the presence of cirrhosis

in daily clinical practice are usually based on e.g. a decrease on platelet counts, increased transaminases, and/or nodular liver surface by imaging methods. However, the diagnostic accuracy of such non-invasive criteria has not been fully determined for the NASH-cirrhosis population[45] and applicants are advised to submit relevant substantiations/justifications for scientific advice in case such a "non-invasively" diagnosed population is intended to be included. Usually, a relevant diagnostic set-up with sufficient validation data available in NASH cirrhosis populations will be required. In the compensated cirrhosis population, adequate criteria to rule out (previous) decompensation are also needed.

In case the proposed endpoint includes a threshold for MELD, an appropriate inclusion criterion for the MELD at baseline will need to be set up that allows measuring relevant deterioration.

5.2.5. Decompensated cirrhotic NASH

Patients with decompensated cirrhosis represent a particularly vulnerable subset of patients. A relevant amount of mechanistic, as well as clinical efficacy and safety data on an investigational compound may be required before the inclusion of such patients into clinical studies. Decompensated cirrhosis could be defined on the (historical) occurrence of at least one "decompensation event" such as variceal haemorrhage, ascites, or hepatic encephalopathy[46]. However, the disease characteristics of NASH decompensated cirrhosis may no longer be sufficiently specific to NASH, but be rather similar or greatly overlapping with decompensated cirrhosis in other liver diseases[47]. Because it is unclear whether a "disease specific development" in decompensated NASH patients is a sensible strategy, applicants are advised to ask scientific advice before embarking on a development program in decompensated NASH cirrhosis.

5.3. Study design and endpoints

The natural history of NASH is assumed to end with the manifestation of cirrhosis in the liver, and the subsequent development of portal hypertension and its sequelae, and decompensation of liver function, which ultimately results in liver associated death, or liver transplantation. Because NASH is also associated with a multitude of risk factors for cardiovascular disease (hypertension, obesity, atherogenic dyslipidaemia, and type 2 diabetes), a relevant proportion of patients will also be prone to causes of death other than liver related ones, mainly cardiovascular.

The long-term endpoint in clinical studies for NASH should include a combination of all-cause mortality, liver transplantation, and the manifestation of decompensation (MELD score, variceal bleeding, ascites, encephalopathy etc.). However, for both cirrhotic and non-cirrhotic NASH, a strategy with the use of intermediate endpoints may apply (see chapter 5.1 and 5.3.1 and 5.3.2)

5.3.1. Non-cirrhotic NASH (fibrosis stage 2 and 3)

Intermediate endpoint

As mentioned earlier, due to feasibility issues to provide long-term outcomes and the unmet medical need in NASH, an interim evaluation of efficacy, with an overall shorter duration of clinical studies could be acceptable for licensing purposes and intermediate endpoints reasonably predicting the long-term outcome have been advocated.

Acceptable intermediate endpoints would consist of two composite endpoints to be evaluated at the individual patient level:

The resolution of NASH – with the presence of any grade of steatosis, and all of the following: No ballooning, only minimal (grade 1) lobular inflammation and – no worsening of the stage of fibrosis.

The improvement of fibrosis by at least 1 stage without any worsening of NASH (no worsening of ballooning and lobular inflammation, not more than 1 grade increase in steatosis).

Efficacy for both endpoints should be demonstrated in co-primary fashion, meaning that both will have to independently demonstrate a statistically significant and clinically relevant difference to placebo. This requirement is thought to take account of the uncertainties associated with a strategy to account for the long-term outcomes later and the need to conclude on a positive benefit-risk at the time of (conditional) approval.

Studies in non-cirrhotic NASH will have to be continued at/after interim evaluation (and potential regulatory approval) as long-term studies to document the benefit for clinical endpoints.

If an intermediate endpoint strategy (with the aim of conditional approval) is used in compounds not deemed adequate to meet the proposed two co-primary composite endpoints (e.g. based on the mechanism of action, e.g. being anti-fibrotic only, or based on the phase 2 results), but a substantial reduction of disease progression/clinical benefit can be anticipated or deduced from phase 1 and phase 2 studies, potential applicants are advised to seek scientific advice and present their proposal with relevant substantiation based on data.

Confirmatory endpoint

The time to manifestation of long-term outcomes is currently largely unknown, and reasonably sized studies in patients with the earlier stages of disease (such as fibrosis stage 2 and 3) with the primary aim to demonstrate an effect on survival free of liver transplant and decompensation events might be unfeasible. Therefore, efficacy endpoints reflecting a substantial increase in the risk of disease progression (to the events described) are considered acceptable. The histological diagnosis of cirrhosis is therefore considered to be acceptable as part of the long-term endpoints. Similar arguments have been accepted for the "model for End-Stage Liver Disease end-stage liver disease" (MELD) score at or above the threshold of 15 (for further discussion, see 5.3.3). The long-term outcome for the demonstration of efficacy in non-cirrhotic NASH is therefore proposed to be a single composite endpoint with any component of the following: all-cause death, decompensation of liver disease (with a complete listing), (histological) diagnosis of (progression to) liver cirrhosis, and MELD ≥15[48].

Control group

In the absence of approved treatments for NASH, placebo appears to be the only acceptable control treatment for clinical studies. This also applies to the long-term extension phases of the studies after evaluation of the intermediate endpoint.

Risks to study integrity arising from dissemination of results (e.g. also by the conditional approval) as well as protecting the study from increased, and potentially differential dropout (from the two treatment groups) are considered to be of utmost importance. Careful timing of the interim evaluation, filing of the MAA, and conduct of the long-term extension phase is required.

According to the legal requirements (see Chapter 5.1) the plans to generate "comprehensive clinical data" will need to be presented at the time of CMA.

This situation may change when one or more substances have been approved.

5.3.2. Cirrhotic NASH (fibrosis stage 4)

Compensated cirrhosis

As a general rule, the composite endpoint with any of the following events: decompensation events (variceal bleeding, hepatic encephalopathy, ascites), MELD score at or above 15, liver transplantation, and death should be used as primary endpoint in studies in NASH cirrhosis.

However, even in patients with manifest cirrhosis, it is currently unclear whether an endpoint strategy with the evaluation of clinical decompensation events, liver transplantation and death is feasible.

Therefore, a strategy with the use of intermediate endpoints collected at an interim evaluation may also be acceptable in this population.

In case the need to use intermediate endpoints in this population is identified, a reasonable endpoint is the reversal of cirrhosis (e.g., defined as "improvement of liver cirrhosis to non-cirrhotic liver disease (at least one point improvement in fibrosis stage)"). The endpoint would need to exclude the occurrence of any decompensation event, an increase in MELD, as well as a deterioration (or re-occurrence) of features of NASH activity (inflammation, ballooning, and fat) at the same time (Improvement of cirrhosis by at least one fibrosis grade without occurrence of a decompensation event and without deterioration of MELD and NAS-score). At this point of time, however, the data available to demonstrate that reversed cirrhosis does indeed also reverse or influence the final prognosis substantially, is considerably less profound than the association shown for progressing disease. Nevertheless, newer data available do tend to confirm this[49]. In case such a study is proposed, potential applicants are advised to substantiate the claim that the prognosis of reversed cirrhosis is similar to the prognosis of (untreated) earlier stages of fibrosis in progressive disease (e.g., from other disease areas such as chronic infectious liver disease, i.e., hepatitis C or B). Moreover, this endpoint will need to be backed concordantly by additional, secondary outcomes, based on histology (e.g., total NAS and components of the NAS), non-invasive markers of disease (imaging techniques, determination of liver stiffness, biomarkers) as well as the available (descriptive) data on decompensation events, liver transplantation, and death.

In case such an intermediate endpoint is used, studies should be continued in order to confirm efficacy based on the clinical long-term outcomes like decompensation events, liver transplantation, and death (=the long-term outcome observation).

The need for providing these outcome data will be assessed based on the proposed overall substantiation of the clinical usefulness of the primary endpoint used and the data on the secondary outcomes. Scientific advice is recommended before planning a study without follow-up beyond the intermediate endpoint.

Control group

Similar to the non-cirrhotic population, in the absence of approved treatments for NASH cirrhosis placebo appears to be the only acceptable control treatment for clinical studies. This also applies to the long-term extension phases of the studies after evaluation of the intermediate endpoint (in case this is used). Concerns with regard to integrity of the study, as well as retention of patients in the study after interim also apply in this population.

Similar to the non-cirrhotic population, the situation may change when one or more substances have been approved.

Decompensated cirrhosis

See Chapter 5.2.5[50].

Developments for non-cirrhotic as well as cirrhotic NASH:

In case applicants intend to develop treatments for the full spectrum of the disease, it is usually expected that at least one study in each of the sub-populations (cirrhotic and non-cirrhotic NASH) is presented. The mentioned features of these studies as given above would need to be considered. Different strategies with regard to completion of studies and intention for intermediate endpoint evaluations (and potential filing for CMA) may need consideration. Regulatory and/or scientific advice may be advisable.

A "mix of disease stages" has also been proposed with the concept of compensated Advanced Liver Disease (cACLD) which emerged from the intention to define an advanced liver disease population in the absence of symptoms and/or clinical signs, but being at high risk of future liver-related morbidity and mortality by the Baveno (VI-VII) conferences[5152]. In NASH, this concept refers in principle to patients with fibrosis stages 3 and 4 and is intended to identify patients with a high risk of progression to complications while being able to avoid liver biopsy. For this, however, it remains currently unclear how the concept with non-invasive diagnosis can be made congruent to the requirements for NASH. Since the concept is relatively new and not fully compatible with the main concepts displayed in this reflection paper, no further recommendation can be given. Applicants intending to pursue a development based on this concept are therefore advised to seek scientific advice before engaging into phase 2 of the clinical development.

5.3.3. Additional considerations on endpoints

MELD-score

The use of the MELD score as part of the composite endpoint in studies with non-cirrhotic NASH is generally regarded to be acceptable. However, the accuracy of the MELD in patients with NASH cirrhosis, especially in case there is concomitant renal impairment has been questioned, and modified MELD scores (MELD-Na, MELD3) [5354] have been advocated. Potential applicants are advised to justify the score proposed based on data in the NASH population, and to have patients undergo a rigorous adjudication for a MELD-related endpoint in order to exclude any non-liver related aetiology (e.g. renal or heart disease). MELD score has also been used as part of the endpoints in studies with the cirrhotic population, and this could be acceptable based on the fact that a MELD ≥15 is usually synonymous to the qualification for the (listing for) liver transplantation. The threshold 15 is, however, not acceptable in studies in the decompensated population, and a thorough justification may be needed in case MELD is intended as part of the composite endpoint in a study in the decompensated population.

Hepatocellular carcinoma (HCC)

The occurrence of HCC is strongly associated with NASH, even in patients without cirrhosis[55]. Studies in NASH have been proposed (and conducted) including the occurrence of HCC into the composite final endpoint evaluation endpoint. However, whether a compound would be able to influence the pathogenetic cascade for the occurrence of HCC within the required time-line of a clinical study is currently unclear[56]. For the time being, the occurrence of HCC should be evaluated as secondary/exploratory or safety (similar to other cancers) endpoint only, unless the inclusion into the composite can be justified based on data.

Occurrence/presence of oesophageal varices

The genesis and development of oesophageal varices is closely related to the development of increased portal pressure. Oesophageal varices could therefore be regarded to present a potential outcome measure in NASH. However, the varices itself without the presence of signs of high risk for bleeding are not regarded to represent an adequate surrogate for the decompensation event of variceal bleeding. Contrary to presence of varices, the occurrence of variceal bleeding itself is a strong prognostic factor, and therefore acceptable as part of the clinical outcomes. Presence, and evaluation of diameter and stigma features of oesophageal varices are not recommended to be part of a primary composite, neither for studies the non-cirrhotic (for the clinical endpoint evaluation), nor for the cirrhotic population.

Hepatic venous pressure gradient (HVPG)

For the grading of portal hypertension, HVPG [57] is considered the gold standard to predict outcomes in patients with compensated and early decompensated cirrhosis of different aetiologies, including NASH. Some data in NASH even suggest that HVPG improvement correlates with overall improved clinical outcome[58]. However, HVPG is invasive and measurement challenging to conduct, and therefore rather regarded to be suitable as a pharmacodynamic marker in early studies with limited number of patients[59]. The endpoint is therefore not considered appropriate as part of the clinical endpoints in the cirrhotic NASH population but may be useful for compounds affecting haemodynamics.

Patient reported outcomes

Within the last several years, it has been detected that NASH, while not associated with specific symptoms, is burdened with a relevant impairment of quality of life, and with unspecific, liver- related and liver unrelated symptoms[6061] particularly in terms of physical functioning, pruritus and fatigue, with deterioration of physical and mental health as NASH progresses[62]. However, in NASH, it is usually difficult to attribute the symptoms to NASH only, since relevant co-morbidities are expected to be present, which might be the reason for the symptoms. Nevertheless, it is recommended to include the evaluation of symptoms and health-related quality of life within clinical studies in NASH with adequate and thoroughly validated questionnaires, of which several have been in development in recent years. Changes in symptoms, as well as health-related quality of life are to be regarded to present rather secondary, if not exploratory outcomes at this time-point. A claim for the symptomatic treatment of the disease independent from disease modifying effects is currently not considered appropriate.

Duration of studies

The currently published phase 2 data for substances under development have mostly evaluated parts of the above proposed endpoints only and the phase 3 studies published have been largely unsuccessful for the evaluation of the intermediate endpoint based on histology[6364]. Therefore, uncertainty exists with regard to the appropriate duration of studies both in terms of the time needed for interim evaluation with the intermediate endpoints, as well as for the time needed to show relevant effects on the long-term composite endpoint. As a general rule, a two-year interim evaluation is recommended, which can be modified based on phase 2 data, the size of the study, patient characteristics, and the requirements with regard to statistical rigor. The final evaluation, as well as the evaluation in the compensated (in case an intermediate endpoint is not used) and decompensated cirrhosis population would be expected to be usually planned with an event-driven evaluation, and therefore, a fixed duration may not be appropriate.

5.3.4. Methodological considerations including estimands

One confirmatory study

The conduct of only one confirmatory study has been suggested for development programmes in NASH. However, potential applicants should be aware on the necessary implications for the need for high quality data with regard to internal and external validity (e.g., consistency across subgroups and endpoints; relevant patient populations evaluated based on clinically relevant endpoints) as well as increased statistical rigour (e.g. a two-sided alpha considerably smaller than 5%). In case a strategy with interim analysis (and potential conditional approval) is pursued, confirmatory tests are required at interim and final analysis and an appropriate multiple testing strategy is needed. Different strategies are possible including a split of the (overall tighter than usual) alpha with "recycling" of the alpha spent for interim analysis or hierarchical with closing of the study if not successful at interim (See: Points to consider on application with 1. Meta-Analyses; 2. One pivotal study. CPMP/EWP/2330/99).

In case it is intended to conduct more than one confirmatory study, proposals for different patient populations with different (fibrosis) stages of the disease included in each of the studies have been suggested. Such a strategy is generally considered acceptable for the intent to obtain an indication for the full spectrum of NASH, due to the fact that a relevant "disease continuum" can be assumed for the NASH population. Studies will normally be regarded as mutually supportive in case consistent results can be demonstrated. However, in certain cases, the details of such an approach may need to be discussed more thoroughly within a scientific advice.

Target of estimation (estimand)

The scientific question(s) of interest, i.e. what the study seeks to address and ultimately, the target of estimation (estimand) should be specified in all its attributes. The study planning, design, conduct, analysis and interpretation must be aligned with the estimand. It is referred to ICH E9(R1) Addendum on estimands and Sensitivity Analysis in Clinical Trials (EMA/CHMP/ICH/436221/2017).

In order to determine the appropriate strategy for a study in NASH, all potential intercurrent events with regard to the clinical studies objectives should be considered. Relevant intercurrent events expected are those associated with almost all clinical studies, such as treatment discontinuation and use of additional medication. Contrary to other fields of development, the use of rescue medication may – for the time being – not be relevant because no specific treatments are available, but could become more relevant in the future. However, a change in background treatment (including significant life-style changes with weight loss, or uptake of relevant alcohol intake) may relevantly affect the outcome. The impact of changes in background treatment and use of rescue medication on the main study endpoints should be evaluated.

For the evaluation of the intermediate endpoint, the outcome regardless of the occurrence of intercurrent events is generally of primary interest (i.e. a treatment policy strategy discussed in the addendum). However, liver-related events or death should be accounted for by a composite strategy.

For the evaluation of the long-term "final endpoint" (potentially the only endpoint in the cirrhotic population), the outcome regardless of occurrence of intercurrent events (i.e. treatment policy strategy) is also of primary interest.

Design and *statistical* analysis

Choices made regarding design and statistical analysis, including the handling of missing data, must be made considering the target of estimation. Therefore, in alignment with the recommended treatment

policy strategy, data with regard to the outcomes of interest should be collected independently from the occurrence of an intercurrent event. Data that is nevertheless not collected, for example in case the endpoint is based on liver biopsy and the biopsy is missing or not evaluable, results in a missing data problem with regard to subsequent statistical inference. Generally, sensitivity analyses to support the robustness of the primary analysis should be provided.

Evaluation of the "intermediate endpoint"

Considering a patient with missing data as a non-responder usually results in a conservative estimate of the treatment effect for the recommended primary estimand. However, as this is a single imputation method, it is unclear what the impact is on operating characteristics of the analyses (particularly type 1 error) due to not accounting for the uncertainty about the imputed values. Therefore, alternative approaches could be considered. However, if missing data occurs after an intercurrent event that is intended to be handled by the treatment policy strategy, the potential influence of the intercurrent event on the outcome needs to be appropriately reflected in the analysis (e.g., placebo multiple imputation may be reasonable after treatment discontinuation to account for potential loss of effect).

Evaluation of the "final endpoint" (potentially the only endpoint in the cirrhotic population)

Aiming at a complete follow-up for the outcome events is of particular importance as patients that are not completely followed are likely to have a different prognosis than patients who complete the study, implying that censoring such patients is probably informative and leads to bias. Non-performance of a scheduled biopsy during follow-up constitutes a missing data problem; as a biopsy during the study is only scheduled if there is a high likelihood of a cirrhosis (in the non-cirrhotic population, e.g., based on surveillance with non-invasive methods such as fibroscan), an event should be imputed in the primary analysis but sensitivity analyses should be conducted.

5.3.5. Combination treatment

It has been advocated, based on the results of currently available phase 2 studies, and the poor results of the currently available phase 3 studies, that a satisfactory treatment of NASH might only be possible, if new investigational compounds are combined (2 or more substances administered simultaneously), ideally with a combination of two different principles of action, e.g., anti-fibrotic, and anti-inflammatory [6566]. Whereas such a strategy can be followed from a theoretical point of view, potential applicants should move forward carefully with such development programmes in a situation with no established therapies available.

For the development of a combination treatment (and ultimately also a fixed-dose combination medicinal product), the general principles with regard to the demonstration of the contribution of the single-substances to the overall effect and the demonstration of the superiority of the combination over its components are applicable (please also refer to the "Guideline on clinical development of fixed combination medicinal products (EMA/CHMP/158268/2017).

The expectations from the regulatory side would be that the combination is based on valid therapeutic principles, but also that for each of the substances involved, the contribution to the therapeutic effect is demonstrated.

This usually involves the exploration of dose-response relationship for the single substances, as well as the combination itself, which is usually addressed with a so-called "factorial design" study in phase 2 of the development. Potential applicants have raised the concern that the conduct of "full factorial design" studies might relevantly delay the development of successful treatments in the field. While this is acknowledged, at least a "restricted" dose-response exploration of the combination partners and of the combination itself will normally be required. Delay of development and the danger of using ineffective

combinations/doses need to be weighed against each other. Any reduction of the exploration of the full dose range will need to be justified.

The demonstration of the contribution to the overall therapeutic effect usually involves the demonstration of the superiority of the combination over the single substances in clinically relevant outcomes in confirmatory manner. Normally, it is therefore expected that this is part of the phase 3 study (and, e.g., be based on at least the intermediate histology endpoints in non-cirrhotic NASH). In case the justification of the combination is intended to be based on earlier (e.g. phase 2) data or on other endpoints than histology, applicants are advised to seek scientific advice.

Normally, the properties of the single substances should be fully explored and described before a combination treatment is developed, but due to the unmet medical need it can also be evaluated during the development of the combination treatment in case use as single substance is not intended.

A combination treatment is normally expected to be developed as either as a second line treatment in patients with insufficient response to mono-therapy, or in patient groups with a very high risk of progression. However, the ongoing unmet medical need and the failure of several therapies in this field may allow an "initial" combination treatment without the identification of very high-risk subgroups. A strong support/rationale with regard to e.g. mechanism of action/biological plausibility, as well as strong phase 2 data are expected in such cases. Also, an adequate justification for the choice of the patient population, the pre-treatment received, and the overall clinical context is expected.

5.4. Safety considerations

General safety requirements will apply to studies in chronic liver diseases, similar to other fields of drug development. The general requirements to focus on the known pharmacodynamic effects, including off-target effects known from early development programme will fully apply. The following paragraphs therefore deal with the specifics of safety evaluation with regard to liver in patients with underlying liver disease, and the cardiovascular safety consideration applicable to NASH

5.4.1. Liver safety

The evaluation of liver safety in the field is considered paramount, and at the same time, hampered by the underlying disease process. The underlying liver disease, as well as fluctuations occurring during the course of clinical studies may hamper the evaluation of hepatic safety due to the overlap in accompanying symptoms, as well as the changes in the routine liver safety biomarkers used, such as transaminases, ALP, and bilirubin. The distinction of fluctuation and flare of the underlying disease, from (sub-) clinical liver damage and true drug-induced liver injury (DILI) caused by an investigational agent is therefore the most important feature of the evaluation of liver safety in both disease entities. The distinction of the type of injury pattern, as well as causality assessment (e.g., using the well-established Roussel Uclaf Causality Assessment Method (RUCAM) criteria, or the newly developed RECAM tool (if adequately justified)[67][68], as well as expert adjudication), and the search for and identification of potential Hy's law cases, are necessary parts of the evaluation of liver safety and potential DILI in clinical studies. In addition, biopsies should be undertaken whenever possible for causality assessment[69][70][71].

Although a generally increased risk of DILI in patients with underlying liver disease is controversial[72] and may depend on the underlying disease[73], in addition to these general requirements a need exists to define different rules for the safety evaluation before, during, and after clinical studies with underlying liver diseases. These alternative approaches may include inclusion criteria (e.g., limits for increased transaminases) algorithms including interruption, and stopping rules, as well as thresholds to define clinically relevant events and the use of novel statistical approaches specifically developed for

this purpose[74][75]. In addition, the inclusion of experimental biomarkers is highly recommended for studies in patients with underlying liver disease, but the influence of the underlying disease on these markers should be known before they are used to help the assessment of safety. It is recommended that all these methods are implemented in addition to the routine liver safety evaluation.

5.4.2. Cardiovascular safety

Because NASH is associated with the obesity epidemic and is regarded as the liver manifestation of the so-called metabolic syndrome, the patient population included in clinical studies in NASH is at increased risks of cardiovascular disease and related events such as myocardial infarction, stroke, and associated death[76]. The overall risk is modified by the presence of concomitant diseases such as arterial hypertension, diabetes mellitus, severe obesity, and hyperlipidaemia[77][78][79][80].

Therefore, in NASH, the principles of the "reflection paper on assessment of cardiovascular safety profile of medicinal products" (EMA/CHMP/505049/2015), are considered applicable. The need for increased requirements will depend on the mechanism of action, and the pre-clinical data showing potentially detrimental effects with regard to cardiovascular safety. Long-term clinical studies in the field are needed to draw a final conclusion. The number of participants and study duration in NASH are expected to be sufficient to address cardiovascular safety in appropriate manner.

It is necessary, not only to focus the safety evaluation on the occurrence of the so-called major cardiovascular events (MACE) but also on the off-target effects of the potential investigational products on parameters potentially influencing the overall cardiovascular risk, such as cholesterol, glucose homeostasis, and (systemic) inflammation. Implementation of (safety) adjudication panels for relevant cardiovascular events is recommended.

Cardiovascular safety documentation will also need to be part of the Risk Management Plan.

5.5. Children and adolescents

5.5.1. NASH in the paediatric population

Similar to other aspects of the obesity and, "metabolic syndrome" epidemic, NAFLD and NASH have been identified to present an increasingly significant health burden in children and adolescents. The prevalence of NAFLD in children is estimated to be around 3-12% depending on age, but in obese population can be as high as 85%. Whereas 2-4 year-old children are expected to suffer from NAFLD at only very low rates, the prevalence in adolescents almost reaches adult levels[81][82][83][84].

Paediatric NASH shares many features of adult NASH with common underlying pathophysiology represented by progression of steatosis with inflammation and fibrosis. Assuming a similar rate of patients developing NASH from the presence of NAFLD as in adults [85], it is clear that NASH is a relevant health problem also in the young age group, although the development of late-stage disease may take years and might be expected to manifest not before reaching adulthood. However, rapid progression to advanced liver disease in childhood has been described[86]. Although paediatric NASH is variable, in the adolescent population the course of NAFLD/NASH is expected to be similar to adults. There are no relevant data for the younger population. Data about natural history in paediatric patients are needed to assess disease evolution and progression[87] However, there are currently no authorised medicinal products in NASH for children and there is a medical need to develop treatments also in this patient population.

As outlined above, the diagnosis of NASH is currently considered to require the conduct of liver biopsy with histological evaluation, and the conduct of clinical studies should be mainly based on repeated

biopsy results. The diagnosis itself is also based on histology in childhood/adolescence patients[8889]. However, the conduct of repeated biopsies in clinical studies is even more associated with ethical and procedural problems when children are concerned, and the need for non-invasive outcomes in this population is therefore considered to be of even higher priority (see also Chapter 5.1).

Furthermore, the histology evaluations available have shown distinct features of paediatric NASH as compared to adults, with the presence of a relevant proportion of patients developing a unique histology with presence of portal-based chronic inflammation (and fibrosis as opposed to the lobular inflammation found in adults and less ballooning[90]). The clinical meaning of this distinct type of histology in children is currently unknown, and consequently, a different histological scoring system may be needed for the paediatric population.

The development of new medicinal products for the treatment of NASH in children therefore requires first of all the collection of new and evaluation of existing data with regard to the natural history of the disease.

5.5.2. Development in paediatric NASH

Drug development in children will require determination of the adequate age range to be studied. Young children (e.g., below 6-10 years) might still be early in the disease process, and therefore be appropriate candidates for non-pharmacological interventions, such as lifestyle and dietary changes, of which success rates (with regard to weight loss) are usually higher than in adults. In addition, pharmacological treatment (off label) or bariatric surgery (when indicated) may limit disease progression. Consequently, the potential for regression of inflammatory changes is similarly considered to be higher[91].

Since data on the natural history of paediatric NAFLD, although limited, suggest an early onset and more aggressive phenotype of the disease compared to adults [92] , pharmacological treatment options are relevant for paediatric NASH. The development of new medicinal products for NASH in children would also need a determination of the quantity of data needed to be available for adults, before conducting therapeutic studies in paediatric subjects. At this point of time it is recommended that relevant clinical studies are deferred until sufficient efficacy and safety data in adults are available.

The availability of further data on natural history, as well as on the individual new compound in adults might already enable to more precisely determine the level of extrapolation that can be applied (see draft: Reflection paper on the use of extrapolation in the development of medicines for paediatrics. EMA/199678/2016) and the type and amount of data that need to be generated in the paediatric population. A distinction between adolescents and children may become relevant. Older adolescents may be included into adult studies if adequately justified.

Generally, the investigation of the appropriate dose (under full consideration of the potential differences in pharmacokinetics in obese and NASH adolescents compared to adults) will be necessary, and the development of age-appropriate formulations as appropriate.

Placebo-controlled studies may still be required, depending on the questions that remain to be answered, and may need to include liver biopsies. The decision on the need for, as well as the conduct of studies with histology endpoints also needs to take full account of the potential for the ethical problems associated with any more than minimally invasive procedures, and may need a careful approach with regard to the patient selection (e.g. selection of age groups, stage and severity of disease, etc.).

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