Review article

From nonalcoholic fatty liver disease (NAFLD) to metabolic dysfunction-associated fatty liver disease (MAFLD) – new terminology in pediatric patients as a step in good scientific direction?

Marta Flisiak-Jackiewicz*, Anna Bobrus-Chociej, Natalia Wasilewska, Dariusz Marek Lebensztejn

Department of Pediatrics, Gastroenterology, Hepatology, Nutrition and Allergology, Medical University of Bialystok, Poland; pegaz@umb.edu.pl

* Correspondence: Marta Flisiak-Jackiewicz M.D., Waszyngtona St. 17, 15-274 Bialystok, Poland, tel.+48607567895, e-mail: m_flisiak@op.pl

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Abstract: Nonalcoholic fatty liver disease (NAFLD) is the most common chronic liver disease in the world, which predispose to more serious hepatic conditions. It ranges from simple liver steatosis to nonalcoholic steatohepatitis (NASH), which may progress to cirrhosis and even end-stage liver disease. Since obesity became one of the most important health concerns wordwide, a considerable increase in the prevalance of NAFLD and other metabolic implications has been observed, both in adults, and children. Due to the coexistence of visceral obesity, insulin resistance, dyslipidemia, NAFLD is considered to be the hepatic manifestation of metabolic syndrome (MetS). These relationship between NAFLD and MetS led to set up in adults new term combining both of these conditions, called metabolic dysfunction-associated fatty liver disease (MAFLD). Based of these findings, we propose set of criteria, which may be useful to diagnose MAFLD in children and adolescents.

Keywords: MAFLD, NAFLD, fatty liver, metabolic syndrome, obesity, children, nomenclature

Introduction

As shown by previous research, nonalcoholic fatty liver disease (NAFLD) is one of the most important causes of liver pathology worldwide and in the coming decades it will probably become the leading cause of end-stage liver disease. The prevalence of NAFLD ranging from 9-37 % in the general population and has significantly increased over the last two decades. The discrepancy in the prevalence of NAFLD among studies is most likely due to differences in diagnostic modalities, accepted standards for the laboratory tests, diagnostic criteria, as well as dietary and lifestyle habits in different regions of the world [1]. Particularly worrying phenomenon is the rising prevalence of NAFLD observed among children and adolescents, which is an effect of globally dramatic dimensions of obesity remaining one of the most challenging problems in western civilization. The authors of a meta-analysis from 2015 based on 74 clinical trials, estimated the prevalence of NAFLD in obese children and adolescents at 34.2 %, whereas in the

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general pediatric population it was 7,6 % [2]. Unfortunately, it may be associated with early unfavorable metabolic complications and the development of more serious liver conditions in which liver transplantation is the only available treatment option [3].

Based on current knowledge, the pathogenesis of NAFLD is a multifactorial and multi-step process. NAFLD is a clinicopathologic condition characterized by abnormal lipid deposition in the liver defined as steatosis of > 5 % of hepatocytes in liver biopsy or fat fraction > 5.6 % assessed by proton magnetic resonance spectroscopy (^{1}HMRS), in the absence of secondary causes of liver injury and excessive alcohol consumption. In children the diagnostic criteria for NAFLD include steatosis in ultrasonography and abnormal liver tests with exclusion of other liver diseases. Liver biopsy is considered golden-standard in NAFLD diagnosis, but it is not performed routinely due to its invasiveness and high cost. Indications for liver biopsy include uncertain diagnosis, suspected advanced liver disease or before pharmacological or surgical treatment [4]. NAFLD is regarded as a spectrum of hepatic conditions, which ranges from simple steatosis without specific inflammatory changes, through more severe nonalcoholic steatohepatitis (NASH), referring to hepatocyte ballooning degeneration with or without fibrosis, which may progress to cirrhosis with all its consequences e.g. increased risk of developing hepatocellular carcinoma (HCC) and/or hepatic decompensation [5-7].

It is widely accepted that NAFLD is pathogenically a "multiple-hit" disease and a combination of genetic predisposition, the role of lipotoxicity, adipocytokines, altered gut-derived microbiome, as well as environmental factors like high-fat diet, excessive fructose consumption, sleep deprivation or sedentary lifestyle [8-11]. Among established clinical conditions closely associated with NAFLD particular attention is paid to abdominal obesity and other features of the metabolic syndrome (MetS) [12-14], including disturbed glucose metabolism with insulin resistance (IR) [15-17], dyslipidemia [18-20], hypertension [21] and other metabolic disorders connected with increase cardiovascular risk [22]. Due to this connection, NAFLD is widely considered as hepatic manifestation of MetS [23]. The pathogenetic mechanisms explaining the relationships between NAFLD and MetS are not fully understood, however visceral obesity, insulin resistance and subclinical inflammation seem to play the key role in developing both diseases. This strict connection between hepatic steatosis and metabolic dysfunctions led to the creation of a new term for fatty liver accompanied by other components of MetS in adults called metabolic dysfunction-associated fatty liver disease (MAFLD). Therefore, the question arises whether proposed terminology should also be appropriately changed in children and adolescents?

Definition of Metabolic Syndrome in Children and Adolescents

MetS is an essential health problem, that involves a group of factors that together increase a risk of cardiovascular disease (CVD) and is associate with IR and type 2 diabetes mellitus (T2DM). It seems that MetS risk factors can appear at any stage of life from childhood to adulthood, that is why there is a necessity to urgently identify children at increased risk for cardiometabolic comorbidity as early in life as possible, to enable them to initiate preventive treatment before concomitant disease has occurred.

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To assess cardiometabolic risk in obese children and adolescents it is important to perform a detailed clinical examination with evaluation of anthropometric measures such as height, weight, waist, and hip circumferences, applying age- and gender-specific centiles. Many studies have shown that waist circumference (WC), waist-hip ratio (WHR), or waist-to-height-ratio (WHtR) may be an useful and simple tools for central obesity screening, as well as cardiovascular risk and NAFLD assessment in both, children and adults [24-26]. The degree of overweight or obesity could be determined based on the calculation of body mass index (BMI), which should be referred to appropriate centiles curves [27]. In a population-based study, it has been shown that adolescents with a BMI ≥ 99th percentile have a significantly greater risk for having cardiovascular risk factor clustering compared to those with lower degrees of obesity [28].

While the definition of MetS in adults is well-establish, it is more problematic in children because of a multitude of criteria with various settings of cut-off values, as well as difficulties in predicting future risk for developing CVD and T2DM. Reinehr et al. compared different MetS definitions in a cohort of 1205 children and adolescents to establish the prevalence of MetS. Researchers emphasized that it differed significantly depending on the definition criteria applied and ranged from 6 to 39% [29]. Some authors suggest that the prevalence of individual components of MetS in the pediatric population would be more relevant for the assessment of cardiovascular risk than the diagnosis of MetS [30]. In 2005, Alberti et al. proposed a global definition for MetS in adults. Diagnostic criteria for MetS in adults include central obesity (increased waist circumference ≥ 94 cm for males and ≥ 80 cm for females) and the presence of at least two additional abnormalities: dyslipidemia (increased triglycerides concentration (≥ 1.7 mmol/l (150 mg/dl) or reduced high-density lipoprotein cholesterol (HDL-C) concentration (< 1.03 mmol/l (40 mg/dl) in males and < 1.29 mmol/l (50 mg/dl) in females) or specific treatment for these lipid abnormalities), high blood pressure (systolic: ≥ 130 mmHg or diastolic: ≥ 85 mmHg or treatment of previously diagnosed hypertension) and glucose intolerance (fasting plasma glucose ≥ 5.6 mmol/l (100 mg/dl) or previously diagnosed 2TDM) [31]. Two years later, a new definition from the International Diabetes Federation (IDF) was developed that included all previous studies [32]. According to the presented criteria in children older than 16 years, the IDF adult criteria may be used for MetS diagnosis. In the group of patients aged 10-16 years diagnosing the MetS requires the presence of abdominal obesity diagnosed based on age- and gender-specific percentile curves of WC (≥90th percentile) and two or more other following metabolic factors: hypertriglyceridemia, low HDL-C, high blood pressure or glucose intolerance. In the youngest children between 6 and 10 years old MetS cannot be diagnosed, but if the child's waist circumference is over or equal to the 90th percentile further measurements should be made especially if there is a family history of metabolic disturbances. However, one of the most important limitations of the proposed MetS definition in children and adolescents is not taking into consideration NAFLD, hyperuricemia or sleep apnea, which are significantly associated with increased cardiometabolic risk in later life. Furthermore, it has been described that many of the metabolic and cardiovascular complications of obesity were already detectable in prepubertal children [33]. Therefore, MetS definition

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should be extended to prepubertal children, which is not currently the case. Due to the possible early onset of metabolic disorders, early prevention of MetS during childhood might not only decrease chronic disease burden early in life but also lower the proportion of adults who will develop cardiometabolic disease. Appropriate therapy and lifestyle change in these patients and their families will help to prevent the negative effects of obesity in the future.

Pathogenetic mechanisms linking NAFLD and MetS in obese children, adolescents, and adults

The relation between NAFLD and MetS is one of the most often discussed problems. Numerous studies have shown a link between them, as well as a strong association of NAFLD in obese children with multiple cardiovascular risk factors (Figure 1). The presence of metabolic disorders such as T2DM, obesity, dyslipidemia, and hypertension carries a high risk of disease progression and development of NASH and fibrosis in NAFLD patients [34-35]. The estimated prevalence of MetS was approximately 18% of non-obese and 67% of obese individuals with NAFLD [6].

The study conducted in overweight and obese children presenting with hepatomegaly and/or elevated alanine aminotransferase (ALT) has revealed that patients labeled as having MetS had significantly higher risk to have NAFLD proven by biopsy than those who didn't meet the criteria of MetS. However, they didn't demonstrate a difference in ALT activity or echogenicity of the liver by ultrasound examination (USG) [36]. Yi-Wen Ting et al. have proven that obese children with MetS are more likely to have advanced liver fibrosis measured by transient elastography (using Fibroscan). Moreover, metabolic components such as BMI, WC, systolic blood pressure (SBP), serum triglycerides (TG) levels, serum total cholesterol (TC) level, hemoglobin A1c (HbA1c), 2TDM, Homeostatic Model Assessment for Insulin Resistance (HOMA-IR) were significantly higher in patients with liver fibrosis. Multivariate regression analysis shown that waist circumference is a significant independent predictor of liver fibrosis [37]. In several studies it was also documented that elevated waist circumference or BMI is strongly correlated to parameters of hepatocyte injury – ALT and gammaglutamyl transferase (GGT) [27]. According to the recent scientific data, obese children with NAFLD diagnosed by both elevated serum ALT activity and liver steatosis on USG, have significantly higher BMI, WC values, ALT and GGT activities, HOMA-IR and intensity of the hepatic steatosis in USG and intrahepatic lipid content in ^{1}HMRS compared to other obese children without NAFLD [38]. Some studies also suggest that liver fat accumulation is a sensitive and early indicator of metabolic dysfunction [39]. This is in line with results from the study of Papandreou et al., who demonstrated that the majority of children with hepatic steatosis assessed by USG fulfilled three or more criteria of MetS (58,6%) and showed significantly higher BMI, WC, TG and lower HDL-C levels, as well as higher fasting insulin levels and higher IR assessed based on HOMA-IR, compared to patients with normal echogenicity of the liver. Moreover, in logistic regression model, patients presenting with MetS have a three times higher risk of developing NAFLD compared to those without MetS [40]. Results from all these studies could be the reason for the necessity

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of undertaking routine control studies toward NAFLD in all obese patients and early lifestyle modifications to effectively reducing the overall risk of MetS. Also in a recent study performed by Prokopowicz et al., a group of obese children with NAFLD was more likely to present MetS than those without NAFLD (40.82% versus 22.81%). NAFLD patients were characterized by greater WC, WHR, significantly higher serum concentration of TC, TG, fasting insulin, as well as glucose and insulin in 120 minutes of Oral *Glucose Tolerance Test* (OGTT). On the other hand, among the MetS components only hypertriglyceridemia was most often recognized in NAFLD patients than in those without NAFLD. Values of other components didn't differ significantly in these group. Important here is the fact that only fasting glucose, but not insulin resistance, is considered in the IDF criteria for MetS. In these study, NAFLD was significantly more often diagnosed in patients with HOMA-IR exceeding reference values than in children with the normal range of HOMA-IR (79% versus 28% for HOMA-IR > 90 percentile; 85% versus 15% for HOMA-IR > 97 percentile). According to the authors HOMA-IR > 4.089 is a good indicator of NAFLD (AUROC = 0.817, sensitivity = 70.8%, specificity = 83.6%) [41].

It has been proven that IR, which is described as a reduced effect of insulin in its target tissue, is increased in obese patients. Within the liver this will be manifested by reduced suppression of hepatic glucose production along with an increase of de-novo lipogenesis and very-low-density lipoprotein (VLDL) production. Normative values of IR as estimated by HOMA-IR calculated from fasting glucose and insulin concentrations. Diagnostic cutoffs of cardiometabolic risk factors in the pediatric population have been proposed by Shashaj et.al, who also included separate standards for obese children. Authors of this study observed that HOMA-IR > 3,42 in obese children is associated with an increased cardiometabolic risk, defined as a presence of at least one of the following: hypercholesterolemia, hypertriglyceridemia, reduced HDL-C levels, and ALT ≥ 40 U/l [42]. Patton et al., by recruiting 254 children and adolescents aged 6–17 years, have demonstrated that the severity of IR was significantly correlated with histological features of NAFLD and the risk of MetS was greater among those with severe steatosis [43]. While, Newton KP et al. evaluated the prevalence of T2DM and prediabetes in a large multi-center cohort of children with biopsy-proven NAFLD from pediatric centers across the United States. The researchers found that nearly 30% of children with NAFLD had abnormal glucose metabolism with 6.5% fulfill the criteria for T2DM. Notably, independent of age and BMI, girls with NAFLD were more likely to have T2DM than boys with NAFLD. Moreover, children with T2DM had a greater risk of having NASH (43.2%), regarded as a more progressive form of NAFLD, compared with prediabetes (34.2%) or those with normal glucose (22%) [44]. Similar observations were included in other studies, where T2DM was associated with more advanced forms of NAFLD within the studied pediatric and adult population [45-46]. Therefore, as one would expect, early detection of liver steatosis in high-risk populations is important for avoiding further development of severe forms of NAFLD. A recent study performed by Koutny et al. included obese patients aged 2-20 years from 51 centers, aimed to determine the prevalence of prediabetes and T2DM in patients with the increased and normal activity of ALT. They demonstrated that obese patients with mild (>24 to ≤50 U/L) or significant (>50 U/L) increase of ALT had

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greater odds ratios for prediabetes, while those with a significant increase of ALT also for T2DM compared to obese controls with normal liver transaminases activity. These findings may suggest that youth with NAFLD have a substantially higher risk of T2DM than obese youth in general [47].

As shown, IR represents a key factor linking the development of MetS features in children with liver steatosis. The strict connection between fatty liver and impaired insulin sensitivity has been documented, but considering the complexity of links between NAFLD, IR, and T2DM, it is extremely difficult to find out whether NAFLD is the effect or the cause of IR. For many years, NAFLD has been treated as the hepatic consequence of peripheral IR, which may affect the liver by different mechanisms [48]. It is most often explained by promoting intrahepatic lipid accumulation by up-regulation of free fatty acid (FFA) influx from adipocytes to hepatocytes. Excess of FFA in the portal system comes from accelerated by IR lipolysis of visceral fat and it may lead to fatty liver and MetS in different ways according to the genetic and epigenetic background. Furthermore, the consequence of this condition is upregulation of de novo lipogenesis, an impairment of FFA β-oxidation and oversecretion of VLDL, with a further increase of hepatic lipid accumulation [49-51]. Additionally, this excessive accumulation of lipid in hepatocytes may interfere with the normal insulin signal transduction pathway. It is also well-established that altered production of adipose tissue-derived circulating factors called adipocytokines is also closely associated not only with obesity, but also with NAFLD and MetS [52-56]. The FFA overload in the hepatocytes together with the release of adipokines and proinflammatory cytokines coming from the adipose tissue are responsible for lipotoxicity, with a consequent mitochondrial dysfunction and increase of cytotoxic reactive oxygen species (ROS) production and the development of inflammatory response, predisposing to progressive liver damage [57].

However, recent evidence shows that the relation between NAFLD and components of metabolic syndrome is much more complicated and bi-directional. Based on recent research cannot be excluded also the theory that hepatic steatosis may precede IR and MetS and even be a risk factor for their development, which can further exacerbate NAFLD leading to its progression towards NASH and fibrosis [58-61]. Assuming that NAFLD is the main cause of IR/MetS, it can be expected that therapies targeted at reducing liver fat storage may be successful in coping with the above metabolic disorders. Indeed, multiple studies across the world have revealed that lifestyle modification through changes in diet and physical activity (inducing a weight loss) in obese patients with NAFLD, apart from the beneficial effect on hepatic features, improved also insulin sensitivity and normalize several components of the MetS [62].

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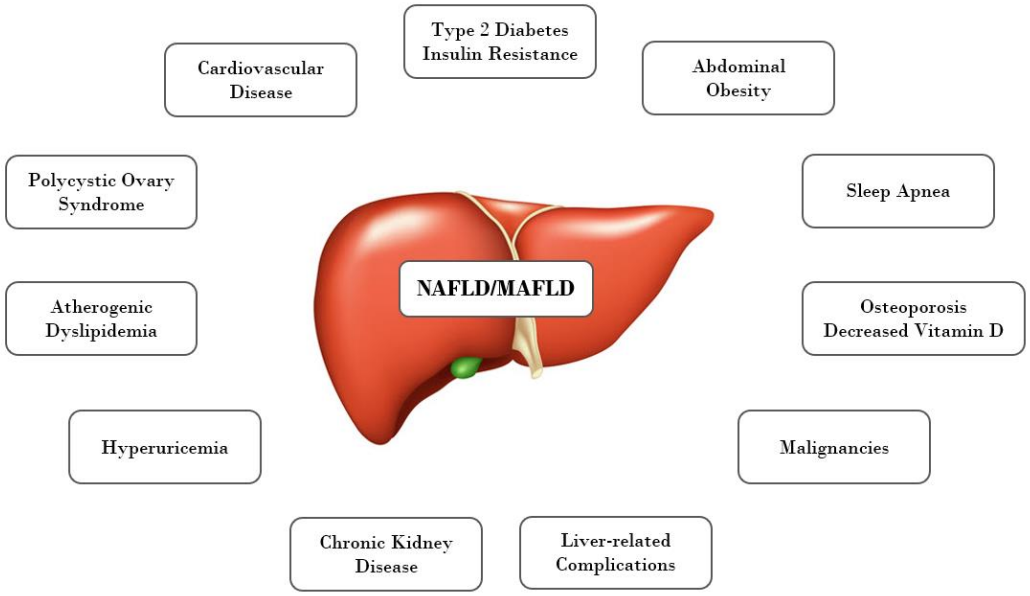

Figure 1. NAFLD/MAFLD: a multi-organ disorder.

From NAFLD to MAFLD

Data obtained from quoted publications suggest that a large part of obese patients with NAFLD present with concomitant metabolic risk factors. Based on this knowledge, recently, panel of experts have proposed the change of the terminology from NAFLD to MAFLD, which strictly correspond with comorbidities [63]. In this document, there are proposed criteria for the diagnosis of MAFLD, which are based on detection of liver steatosis by imaging techniques, blood biomarkers/scores, or by liver histology in addition to one of the following three criteria: overweight/obesity, presence of T2DM or evidence of metabolic dysregulation, characterized by the presence of at least two metabolic risk abnormalities (increased waist circumference, high blood pressure, hyperglyceridemia, reduced HDL-C concentration, glucose intolerance or increase HbA1c, HOMA-IR ≥ 2,5, increased high-sensitivity C-reactive protein (hs-CRP) level in serum).

Recent data have shown that normal body weight doesn't protect from the development of NAFLD and it can also occur in non-obese individuals [64-65]. The incidence of lean NAFLD in adolescents ranging from 8% in the USA to 16% in the Asia-Pacific region [66]. According to MAFLD criteria, we are able to diagnose it also in non-overweight/non-obese patients, when we confirm a presence of liver steatosis with at least two mentioned above metabolic risk abnormalities. It should be noted that these non-obese patients with MAFLD are also at greater risk of liver damage and cardiovascular risk in comparison with metabolically healthy individuals [67].

The current NAFLD definition demands to rule out other causes of fatty liver. A new diagnostic strategy of MALFD, based on inclusion rather than exclusion criteria, accepts that development of fatty liver is associated with "multiple hits" and several different components may interact synergistically with metabolic factors [68-69]. Thus, MAFLD is

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a more specific term that describes more realistically the basis of this disease. This new nomenclature includes only adults so far, however, it could be also used successfully among children and adolescents, just to emphasize that liver steatosis can be present in patients with MetS and IR, independently of age. Hence, we come up with a proposal criteria set for the diagnosis of MAFLD in children and adolescents. In accordance with the IDF consensus report [32], which defines the MetS, MAFLD should not be diagnosed below the age of 10 years as well. In the older children between 10 and 16 years old a diagnosis of MAFLD can be made based on the presence of hepatic steatosis in combination with one of the following three criteria:

1. abdominal obesity assessed by WC ≥ 90th percentile adjusted for age and gender,
2. high fasting plasma glucose > 100 mg/dl or known T2DM or
3. presence of at least two metabolic risk disturbances in lean patients – elevated triglycerides, low HDL-C, high blood pressure, HOMA-IR ≥ 2,5, increased hs-CRP (Figure 2). MAFLD adult criteria can be used for adolescents aged more than 16 years old. The proposed redefinition of the disease has the potential to improve people's awareness about the pathogenesis of this common liver condition and help in understanding multiple metabolic dysfunctions that affect the human body from an early age. This could facilitate the identification of this kind of patient, especially that nowadays there is a huge development of new noninvasive markers useful for the prediction of hepatic steatosis and progression to steatohepatitis and hepatic fibrosis, without the need for routine performing liver biopsy [70]. Finally, earlier diagnosis of fatty liver is synonymous with sooner intervention and fewer complications in future life.

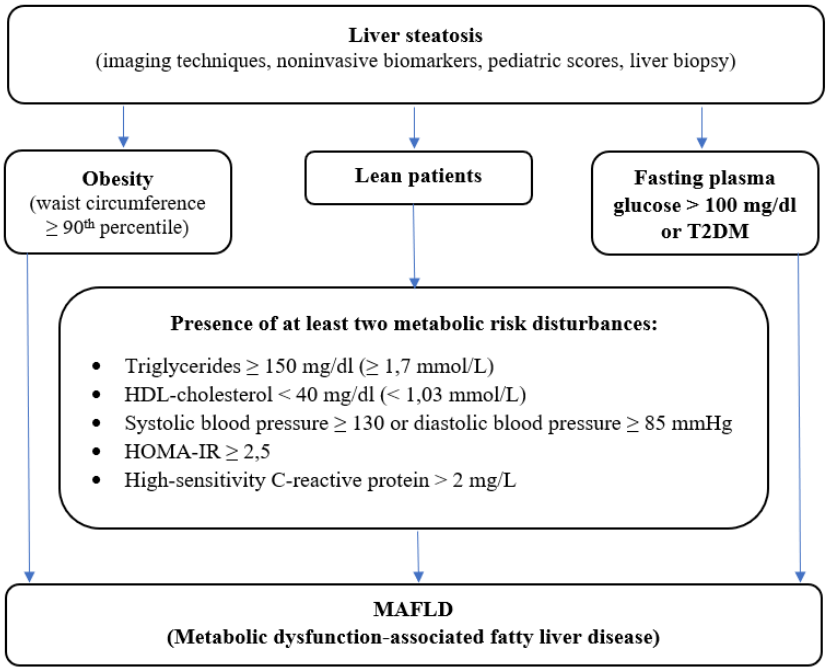

Figure 2. A proposed diagnostic criteria for MAFLD in patients aged 10-16 years old (adopted from Eslam et al. and IDF definition of MetS in children and adolescents)

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