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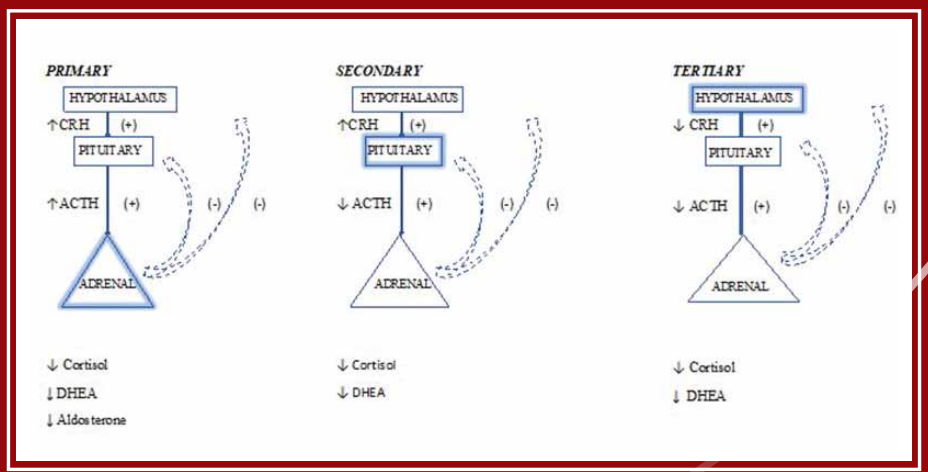

Types of adrenal insufficiency

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ACHAIKI IATRIKI

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CONTENTS

Dear colleagues,

In the current issue, the original research article by Pastras et al. assesses the impact of hepatitis B virus (HBV) infection on non-alcoholic fatty liver disease (NAFLD) patients and evaluates their clinical and laboratory characteristics. In parallel, this original article examines the association of HBV-NAFLD coexistence with liver disease development.

Moreover, this issue includes five review articles. The first review by Mageiropoulou et al. summarizes the latest data regarding the clinical manifestations, evaluation, diagnosis, and current treatment options in thyroid diseases, in adults. The review by Tsiri et al. provides the current knowledge concerning the recently published data on the chronic obstructive pulmonary disease (COPD) characteristics, in order to suggest a therapeutic algorithm. The review by Kalafateli M. focuses on liver complications related to immune checkpoint inhibitors treatment, and discusses the incidence, diagnosis and treatment strategy, currently used in this setting. The review by Psaromyalou et al. presents data on hyperthermia, a medical emergency which can lead to multi-organ dysfunction and even death if left untreated, pointing out the wide differential diagnosis of this condition and the great attention that emergency medicine physicians should provide. Lastly, the review by Armeni et al. provides knowledge regarding pathophysiology, clinical manifestations, underlying aetiologies, and related treatments in adrenal insufficiency.

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ORIGINAL RESEARCH ARTICLE ACHAIKI IATRIKI | 2023; 42(2):57–65

Non-alcoholic fatty liver disease (NAFLD) and hepatitis B virus (HBV) interplay: Their role in liver disease development

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Abstract

Background and Aims: Hepatitis B virus (HBV) infection is associated with lower risk of non-alcoholic fatty liver disease (NAFLD) in the absence of concurrent metabolic disorder. The aim of this retrospective case-control study is to evaluate the impact of HBV infection on NAFLD patients, the clinical/laboratory characteristics of NAFLD-HBV patients and the NAFLD-HBV coexistence relation with liver disease development.

Methods: The medical charts of 575 NAFLD patients referred to outpatient clinic due to abnormal liver biochemistry and/or the presence of fatty liver were thoroughly reviewed. Finally, 518 patients were included in the study; 402 NAFLD and 116 NAFLD-HBV patients.

Results: NAFLD-HBV patients had significantly lower γ-GT, and platelets, and higher ALP and INR compared to NAFLD patients. Lower percentage of NAFLD-HBV patients were overweight/obese compared to NAFLD patients. NAFLD-HBV patients admitted to hospital more often than NAFLD patients; no difference demonstrated in mortality. In multivariate analysis, HBV coexistence, diabetes mellitus, platelet count and total bilirubin were demonstrated as independent prognostic factors for liver disease development.

Conclusions: NAFLD-HBV comorbidity was associated with reduced body weight, increased hospital admissions risk and liver disease development. NAFLD-HBV coexistence constituted an independent risk factor for liver disease development. Thus, active treatment for both disorders should be recommended.

Key words: *Non-alcoholic fatty liver disease (NAFLD); Hepatitis B virus (HBV); Liver disease; Metabolic disorder; Risk factor*

INTRODUCTION

Non-alcoholic fatty liver disease (NAFLD) is one of the most common liver diseases, characterized by several degrees of liver damage, ranging from excessive fat accumulation known as hepatic steatosis, to liver injury and liver inflammation, known as nonalcoholic steatohepatitis (NASH), ending up in advanced fibrosis, and liver cirrhosis [1-4]. NAFLD presents a growing global prevalence of roughly 25% in the general population and is closely associated with high rates of hepatocellular carcinoma (HCC), liver transplantation, and mortality [2-4].

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Hepatitis B virus (HBV) infection constitutes a major public health problem worldwide with an estimated prevalence approximately 3.5% [5]. Despite the widespread use of antiviral drugs and vaccination campaigns, there are approximately 350-400 million HBV patients worldwide, who carry high risk of liver cirrhosis and HCC [2,3,6].

Accompanied by the growing prevalence of NAFLD, over recent years, coexistence of NAFLD and HBV is commonly encountered [7]. The accumulating rate of NAFLD among HBV-infected patients is worrying; it is estimated that 29.6% of HBV patients have NAFLD [8,9]. This comorbidity has attracted widespread attention and interest, focusing on the interaction and the association between these two diseases [10,11]. However, great interest is focused on that although NAFLD and chronic hepatitis B (CHB) can collectively deteriorate liver damage and increase the risk of liver cirrhosis and HCC [3,12-15], NAFLD per se seems to present a positive relationship with decreased HBV seromarkers in HBV patients [16-18]. In parallel, HBV patients also exhibited diminished lipidemia incidence [19-21] and NAFLD onset [7,20,22-24].

Thus, the complex interplay between HBV and NAFLD remains vague and their impact on liver disease course is under investigation. The aim of this study was to assess the impact of HBV infection on NAFLD patients and to evaluate their clinical and laboratory characteristics. Secondary aim of the study was to examine the association of HBV-NAFLD coexistence with liver disease development.

MATERIALS AND METHODS

Study population and selection of patients for analysis

Patients referred to the outpatients' clinic of Division of Gastroenterology in University Hospital of Patras, Greece, due to the presence of NAFLD were enrolled to the current retrospective case-control study. In total, the medical charts of 575 NAFLD patients were initially retrieved. In figure 1, the flow chart for inclusion and exclusion criteria for subjects' enrollment in the current study is presented. Therapeutic and diagnostic criteria were applied constantly during the follow-up period. Patients underwent clinical evaluation in the outpatients' clinic at regular intervals according to the current guidelines [25]. Patients with positive serologic markers for hepatitis C (HCV) (n=26), autoimmune hepatitis autoantibodies (n=10),

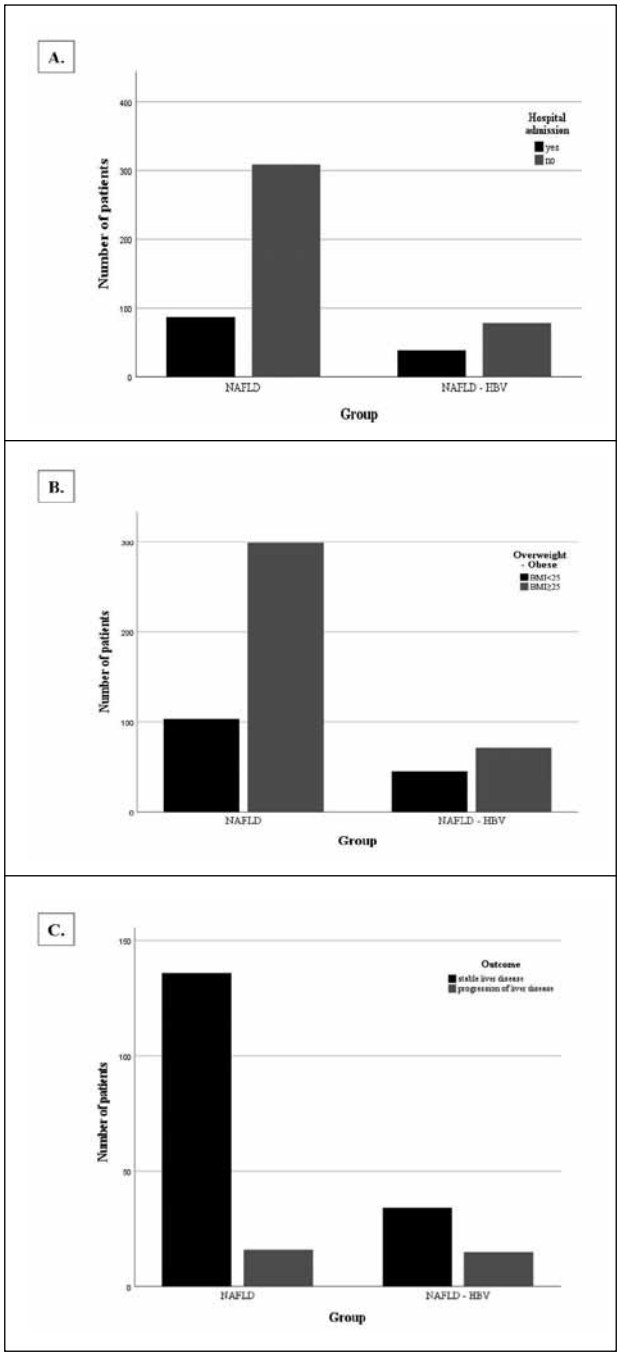

Figure 1. Flow chart of the study design.

primary biliary cirrhosis (PBC) (n=3), hemochromatosis (n=2), human immunodeficiency virus (HIV) (n=1) infection, inflammatory bowel diseases (IBD) (n=3) were excluded from the study [26]. Moreover, excessive alcohol intake (≥210 g/week for men and ≥140 g/week for women) and pregnancy were applied as exclusion criteria [26,27]. Patients who had missing

anthropometry data or metabolic parameters, were not included in this study. Lastly, the presence of liver cirrhosis at baseline was an exclusion criterion, as well. HBV infection alongside NAFLD was reported in 125 patients. Finally, the examined groups of our study comprised 402 individuals with NAFLD and 116 patients with NAFLD and HBV (Figure 1).

Acquisition of clinical data

Participants' demographic data (age, gender, sex), alcohol consumption, smoking, alcohol intake, physical activity, medical history, prior medication use, weight, height, body mass index (BMI), blood pressure, presence of diabetes mellitus, dyslipidemia and hypertension collected from our electronic database. Clinical characteristics were determined according to established criteria [28-31].

Acquisition of laboratory data

Laboratory data were also recorded, including urea, total bilirubin, direct bilirubin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), creatinine, alkaline phosphatase (ALP), glutamyl transpeptidase (γ-GT), hemoglobin, platelets (PLTs), total cholesterol, triglycerides, international normalized ratio (INR), alpha-fetoprotein (aFP), low density lipoprotein cholesterol (LDL-C), high density lipoprotein cholesterol (HDL-C) and fasting blood glucose.

Measurements and definitions

Radiologic information was obtained from ultrasound stiffness imaging methods. NAFLD diagnosis was based on the American Association for the Study of Liver Diseases (AASLD) and European Association for the Study of the Liver (EASL) - European Association for the Study of Diabetes (EASD) - European Association for the Study of Obesity (EASO) Clinical Practice Guidelines for the Management of NAFLD [25,32]. Obesity was defined according to the Clinical Practice Guidelines for the management of adult obesity by the EASO, as underweight when BMI <18.5, normal range when BMI 18.5–24.9, overweight when BMI 25.0–29.9, obesity when BMI ≥30 [33]. Liver cirrhosis diagnosis was based on clinical, histological, laboratory, and ultrasound findings [34,35].

Statistical Analysis

Continuous variables were expressed as medians and interquartile ranges (IQRs), while categorical variables were presented as absolute numbers and corresponding percentages. Categorical data were compared using Pearson's chi-squared test or two-sided Fisher's exact test, when applicable. Regarding the continuous variables, Mann-Whitney U test was used to compare differences between two independent groups. Binary logistic regression was performed to assess the risk factors associated with liver disease progression. Liver disease development was defined as a composite endpoint, namely as progression to cirrhosis or HCC development. First, each variable of interest was included in a univariate model and, subsequently, all variables with a p-value<0.05 were included in the multivariate model. A stepwise approach based on backwards elimination was applied. Statistical analysis was performed using the statistical package IBM SPSS version 26.0. The threshold of statistical significance was set at 5% ($p \leq 0.05$).

Ethics

The study protocol was reviewed and approved by the Ethics committee of the University Hospital of Patras, Patras, Greece. The study protocol conformed to the ethical guidelines of the Declaration of Helsinki for medical research involving human subjects.

RESULTS

Patients' characteristics and comparisons between NAFLD-HBV and NAFLD groups

Patients' clinical, demographic, and biochemical characteristics at baseline are presented in Table 1. Overall, 402 patients with NAFLD and 116 patients with NAFLD and HBV were included. Overweight and obese individuals (BMI>25) were significantly higher on NAFLD group (74.4%) compared to NAFLD-HBV group (61.2%) (p=0.007) (Figure 2A). NAFLD-HBV patients presented significantly lower levels of serum γ-GT (p<0.001) and platelets (p=0.041), whereas ALP levels (p=0.023) and INR (p=0.028) were statistically higher in NAFLD-HBV compared to NAFLD group.

NAFLD *versus* NAFLD-HBV patients' outcomes

Hospital admissions, development of liver cirrhosis, development of HCC and mortality were examined as the final outcomes in our study groups. The results showed that the hospital admissions were significantly higher in the NAFLD-HBV group (35.8%) compared to the NAFLD group (22%) (p=0.004) (Table 2 and Figure

Table 1. *Patients' demographics, main clinical and biochemical characteristics at baseline.*

	NAFLD-HBV		NAFLD		
Variable	N	Percentage (%)	N	Percentage (%)	p-value
Sex (M/F)	67/49	57.8/42.2	208/194	51.7/48.3	0.291
Smoking (yes)	34	29.3%	115	28.6%	0.906
BMI>25 (kg/m²)	71	61.2%	299	74.4%	0.007
Hypertension (yes)	47	40.5%	153	38.1%	0.665
Diabetes Mellitus (yes)	33	28.4%	103	25.6%	0.551
Dyslipidemia (yes)	80	68.9%	293	72.9%	0.413
Variable	Median	IQR	Median	IQR	p-value
Age (years)	53	44.5-63.5	53	42-61	0.746
AST (U/L)	40	24.5-55	36	25-58	0.783
ALT (U/L)	45.5	29-79	54.5	31-81.5	0.208
γ-GT (U/L)	33.5	19-75	55	29-109	<0.001
ALP (U/L)	114.5	87-180	98	70-160	0.023
Total Bilirubin (mg/dL)	0.7	0.52-0.9	0.7	0.5-1.0	0.876
Direct Bilirubin (mg/dL)	0.2	0.105-0.3	0.2	0.11-0.22	0.442
Urea (mg/dL)	33	25-40.5	33	28-39	0.710
Creatinine (mg/dL)	0.9	0.8-1.0	0.9	0.8-1.0	0.862
Fasting glucose (mg/dL)	95	87.5-114	101	92-114	0.060
Cholesterol (mg/dL)	216	191.5-244.5	215	187-245	0.957
LDL-cholesterol (mg/dL)	140	102-156	132.5	109.5-161.3	0.737
HDL-cholesterol (mg/dL)	51	41-60	48	40-58.5	0.572
Triglycerides (mg/dL)	126	93.5-173.5	136	100-187	0.225
Hb (g/dL)	14	13-15	14	13.1-15.3	0.330
Plt (cells/μL)	212	172-265	226	190-270.5	0.041
INR	1.0	1.0-1.14	1.0	0.955-1.065	0.028
aFP (ng/ml)	3.3	2.07-5.27	2.9	2.1-4.3	0.101
Liver Stiffness (kPa)	7.4	6/65-12.05	8.1	5.85-10.15	0.685
Weight (kg)	81	70-93.5	83	72-92	0.522
BMI	28.6	25-31.6	29.1	26.6-31.93	0.176

NAFLD: non-alcoholic fatty liver disease, HBV: hepatitis B virus, N: number of patients, M/F: male/female, IQR: interquartile range, AST: aspartate aminotransferase, ALT: alanine aminotransferase, γ-GT: Gamma-Glutamyl Transferase, ALP: Alkaline phosphatase, LDL: low-density lipoprotein, HDL: LDL high-density lipoprotein, Hb: hemoglobin, Plt: platelets, INR: International normalized ratio, aFP: alpha-fetoprotein, BMI: body mass index

2B). The cumulative mortality rate was 1.7% (2 out of 116 patients) in the NAFLD-HBV group compared to 1.8% (7 out of 400 patients) in the NAFLD group (p=0.999).

Liver disease development

NAFLD-HBV group presented more progressive liver disease compared to NAFLD group (p<0.001). Especially, 20.4% of NAFLD-HBV patients developed liver cirrhosis compared to 7.6% of the NAFLD patients (p=0.011) (table 2). As far as HCC development, 7.4% of the NAFLD-HBV group developed HCC, compared to 1.1% of the NAFLD group (p=0.025) (Table 2). Figure 2C

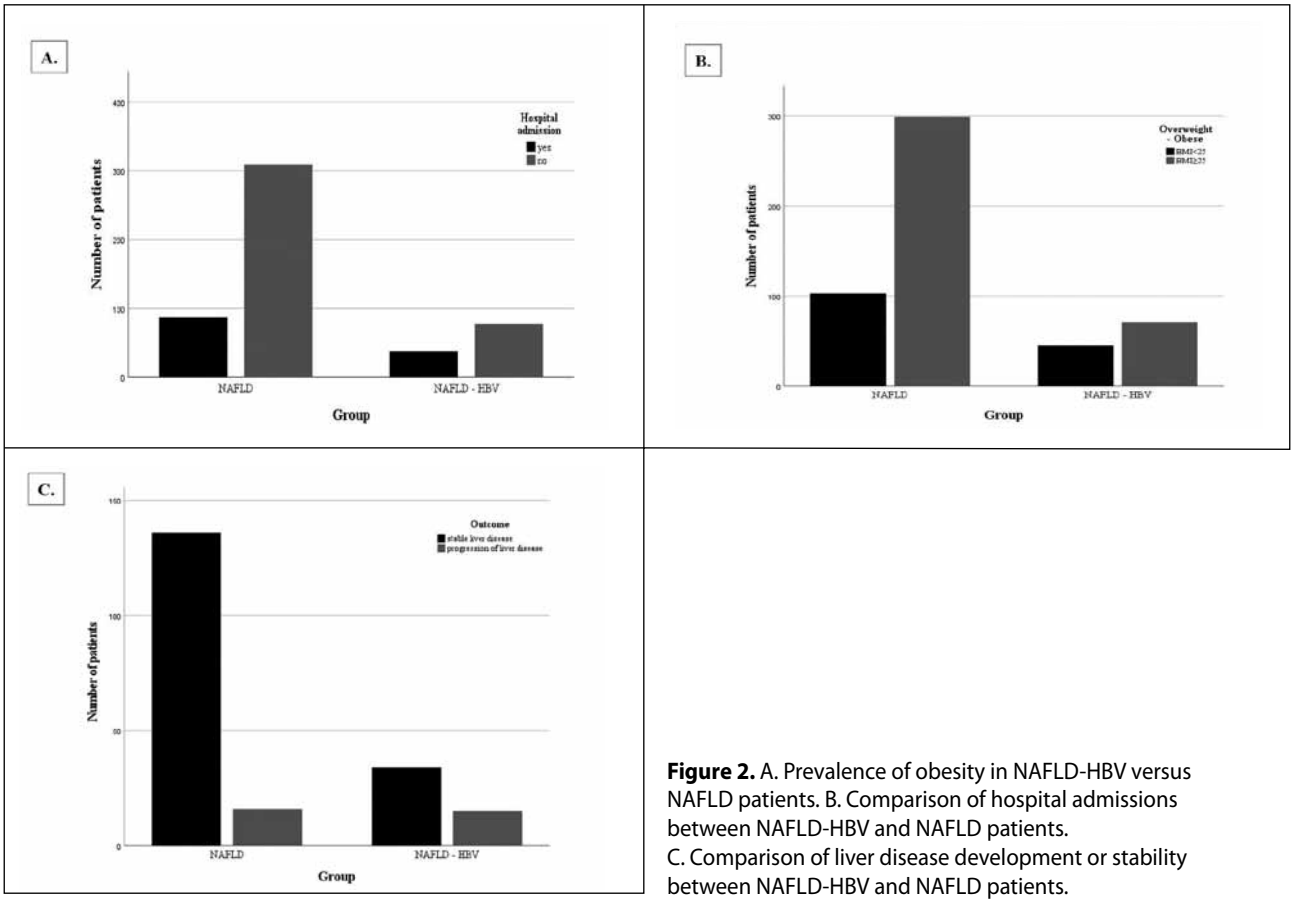

Figure 2. A. Prevalence of obesity in NAFLD-HBV versus NAFLD patients. B. Comparison of hospital admissions between NAFLD-HBV and NAFLD patients. C. Comparison of liver disease development or stability between NAFLD-HBV and NAFLD patients.

presents liver disease development or stability among the examined groups.

Binary regression analyses were applied, exploring the factors associated with liver disease development (progression to cirrhosis or HCC development) (Table 3). HBV coexistence (p=0.001), hemoglobin levels (p=0.014), platelet count (p<0.001), total bilirubin (p=0.003) and the presence of diabetes mellitus (p=0.005) were identified as significant predictors in univariate models. In the multivariate analysis, the HBV coexistence (aOR=3.509, 95% CI: 1.201-10.254, p=0.022), the presence of diabetes mellitus (aOR=3.375, 95% CI: 1.176-9.683, p=0.024), the platelet count (aOR=0.976, 95% CI: 0.965-0.987, p<0.001) and the total bilirubin levels (aOR=1.785, 95% CI: 1.145-2.781, p=0.01) were demonstrated as independent prognostic factors for liver disease development.

Table 2. *Patients' outcomes in NAFLD-HBV versus NAFLD patients*

Patients' outcomes	NAFLD-HBV		NAFLD		p-value
	N	Percentage (%)	N	Percentage (%)	
Hospital admission	38/106	35.8%	87/396	22 %	0.004
HCC development	4/54	7.4%	2/184	1.1%	0.025
Liver cirrhosis development	11/54	20.4%	14/184	7.6%	0.011
Liver disease progression (cirrhosis or HCC development)	15/54	27.8%	16/184	8.7%	<0.001
Death	2/116	1.7%	7/400	1.8%	0.999

NAFLD: non-alcoholic fatty liver disease, HBV: hepatitis B virus, N: number of patients, HCC: hepatocellular carcinoma

Table 3. *Univariate and multivariate analysis of factors associated with risk for disease development (progression to cirrhosis or HCC development).*

Variable	Univariate Analysis	OR (95% CI)	Multivariate Analysis	aOR (95% CI)
Age (years)	0.192	1.021 (0.989-1.055)		
Gender*	0.481	1.321 (0.609-2.865)		
BMI (kg/m²)	0.131	1.057 (0.984-1.135)		
HBV coexistence	0.001	3.75 (1.688-8.332)	0.022	3.509 (1.201-10.254)
Diabetes Mellitus	0.005	3.125 (1.418-6.886)	0.024	3.375 (1.176-9.683)
Hypertension	0.234	1.597 (0.739-3.449)		
Dyslipidemia	0.387	0.685 (0.291-1.614)		
Liver Stiffness	0.055	1.185 (0.974-2.055)		
Hemoglobin (g/dL)	0.014	0.759 (0.61-0.946)	0.166	0.801 (0.585-1.097)
Platelet count (10⁹/L)	<0.001	0.971 (0.96-0.982)	<0.001	0.976 (0.965-0.987)
ALT (IU/L)	0.126	1.002 (0.999-1.005)		
AST (IU/L)	0.061	1.003 (1-1.006)		
γ-GT (IU/L)	0.589	1.001 (0.998-1.004)		
ALP (IU/L)	0.855	0.999 (0.996-1.003)		
Total Bilirubin (mg/dL)	0.003	1.89 (1.238-2.886)	0.01	1.785 (1.145-2.781)
Creatinine (mg/dL)	0.774	1.178 (0.385-3.609)		

OR: odds ratio, aOR: adjusted odds ratio, CI: confidence interval, BMI: body mass index, HBV: hepatitis B virus, ALT: alanine aminotransferase, AST: aspartate aminotransferase, γ-GT: Gamma-glutamyl transferase, ALP: alkaline phosphatase

*Reference category for gender is male.

DISCUSSION

The coexistence of NAFLD and HBV is a topic which has gained increasing interest as both diseases establish abnormalities on the liver histopathology and enzymes that potentiate end-stage liver disease alongside with HCC [2,8,9]. Emerging evidence has surprisingly demonstrated a potential "competitive relationship" between CHB and NAFLD; reduced HBV markers in NAFLD patients and lower risk of NAFLD presence in HBV patients. The current study focuses on the impact of HBV infection on NAFLD patients evaluating their clinical and laboratory parameters and the risk of patients' outcomes and liver disease development.

Obesity, diabetes mellitus and metabolic syndrome (MetS) are independent risk factors for liver cirrhosis and HCC in patients with CHB, proposing a synergistic role of metabolic factors and CHB on HCC pathogenesis [36-38]. Given that NAFLD is the main liver-related manifestation of obesity and metabolic disorders, HBV infection overlapping with NAFLD is likely to further induce the liver cirrhosis and HCC risk.

Our results showed that overweight - obese individuals' rate was significantly higher on NAFLD subjects compared to NAFLD-HBV, a finding that probably explains the reduced rates of comorbidity with NAFLD reported in the literature. However, the biochemical analyses did not reveal a propitious profile in any group; although NAFLD-HBV patients presented abnormal values of ALP, the NAFLD group presented impaired levels of γ-GT. Recent data claimed that HBV infection was associated with lower risk of NAFLD in patients without metabolic disorders. The regional epidemiology and risk factors of NAFLD in CHB patients has been studied extensively in China [11]. A hospital-based study with 14,452 patients indicated that the prevalence of NAFLD ranged from 29.9% to 35.8% in CHB, which was lower in past-infection prevalence rate compared to the general population, as also suggested by prior findings [2,11]. A cross-sectional study of 33,439 Taiwanese subjects displayed an inverse correlation between HBV infection and NAFLD prevalence [22]. Another study using as reference the proton magnetic resonance spectroscopy (MRS), demonstrated a lower prevalence of fatty liver in HBV patients than in patients without HBV [7,20]. Taking all these data into consideration, the current evidence supports that HBV infection could act as a protective shield for NAFLD [17,20,21,39]. However, due to the relatively small sample sizes of the studies, authors concluded that the delineation of the NAFLD role in NAFLD-HBV patients is still far to be reached and further research is needed. Additionally, another point of consideration is that the aforementioned studies did not compare the HBV infected population to the non-infected, restricting their liability and the interpretation of results [2]. Lastly, several data from bedside studies are opposed to the results from basic research regarding the association between HBV infection and NAFLD [17,20,21,39].

One more finding of the current study was that the NAFLD-HBV group was at higher risk of hospital admissions and developing severe complications, including liver cirrhosis and HCC compared to NAFLD group, suggesting that HBV coexistence increases the risk of end-stage liver disease in NAFLD patients. The effect of HBV-NAFLD on liver disease progression remained significant after adjustment for potential confounding factors. These results are consistent with previous studies reporting that NAFLD was an independent risk factor for HCC development in HBV patients whose HBV DNA was suppressed [10]. Other studies reported that concurrent NAFLD augmented the risk of HCC among patients with CHB [3,13]. The effect of NAFLD on HCC development may have additive influence on HBV patients. Despite, HBV affects the NAFLD incidence, the coexistence of NAFLD-HBV may independently augment the HCC risk, which is probably performed by the same mechanism that NAFLD alone promotes HCC.

Some limitations of the current study should be acknowledged. Firstly, this study has retrospective design, and the second limitation concerns its monocentric nature.

In conclusion, our study demonstrated that NAFLD-HBV comorbidity was associated with reduced body weight, increased risk of hospital admissions and end-stage liver disease development. Moreover, although both diseases are well-known to augment the risk of chronic liver diseases and HCC, our study demonstrated that the coexistence of these disorders also constituted an independent critical risk factor for liver disease development. Thus, as NAFLD and CHB deteriorated clinical outcomes, active treatment for both disorders should be recommended.

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Declaration of funding sources: *None to declare*

Author contributions: *P.P., I.A., S.K., A.A., E.T., M.K., V.I., A.B., D.D., E.Z., E.E.K., G.D., and A.T. were responsible for data acquisition and drafting the article; E.T. was responsible for statistical analysis; P.P., S.K., I.A., and E.T. were responsible for data interpretation; C.T. was responsible for study conception and design; K.T. and C.T. were responsible for critical revision of the article for important intellectual content. All authors read and approved the final manuscript.*

REFERENCES

1. Byrne CD, Targher G. NAFLD: a multisystem disease. J Hepatol. 2015;62(1 Suppl):012.
2. Huang J, Jing M, Wang C, Wang M, You S, Lin S, et al. The impact of hepatitis B virus infection status on the prevalence of nonalcoholic fatty liver disease: A population-based study. J Med Virol. 2020;92(8):1191-7.

3. Chan AW, Wong GL, Chan HY, Tong JH, Yu YH, Choi PC, et al. Concurrent fatty liver increases risk of hepatocellular carcinoma among patients with chronic hepatitis B. J Gastroenterol Hepatol. 2017;32(3):667-76.
4. Ma C, Kesarwala AH, Eggert T, Medina-Echeverz J, Kleiner DE, Jin P, et al. NAFLD causes selective CD4(+) T lymphocyte loss and promotes hepatocarcinogenesis. Nature. 2016;531(7593):253-7.
5. Zampino R, Boemio A, Sagnelli C, Alessio L, Adinolfi LE, Sagnelli E, et al. Hepatitis B virus burden in developing countries. World J Gastroenterol. 2015;21(42):11941-53.
6. Lin S, Wang M, Liu Y, Huang J, Wu Y, Zhu Y, et al. Concurrence of HBV infection and non-alcoholic fatty liver disease is associated with higher prevalence of chronic kidney disease. Clin Res Hepatol Gastroenterol. 2021;45(2):6.
7. Wong VW, Wong GL, Chu WC, Chim AM, Ong A, Yeung DK, et al. Hepatitis B virus infection and fatty liver in the general population. J Hepatol. 2012;56(3):533-40.
8. Machado MV, Oliveira AG, Cortez-Pinto H. Hepatic steatosis in hepatitis B virus infected patients: meta-analysis of risk factors and comparison with hepatitis C infected patients. J Gastroenterol Hepatol. 2011;26(9):1361-7.
9. Zhu L, Jiang J, Zhai X, Baecker A, Peng H, Qian J, et al. Hepatitis B virus infection and risk of non-alcoholic fatty liver disease: A population-based cohort study. Liver Int. 2019;39(1):70-80.
10. Cho H, Chang Y, Lee JH, Cho YY, Nam JY, Lee YB, et al. Radiologic Nonalcoholic Fatty Liver Disease Increases the Risk of Hepatocellular Carcinoma in Patients With Suppressed Chronic Hepatitis B. J Clin Gastroenterol. 2020;54(7):633-41.
11. Zheng Y, Xu K, Hu H, Draz MS, Wu W, Li L. Prevalence and Incidence of Non-alcohol Fatty Liver Disease in Chronic Hepatitis B Population in Southeast China: A Community-Based Study. Front Med. 2021;8:683872.
12. Peleg N, Issachar A, Sneh Arbib O, Cohen-Naftaly M, Braun M, Leshno M, et al. Liver steatosis is a strong predictor of mortality and cancer in chronic hepatitis B regardless of viral load. JHEP Rep. 2019;1(1):9-16.
13. Lee YB, Ha Y, Chon YE, Kim MN, Lee JH, Park H, et al. Association between hepatic steatosis and the development of hepatocellular carcinoma in patients with chronic hepatitis B. Clin Mol Hepatol. 2019;25(1):52-64.
14. Charatcharoenwitthaya P, Pongpaibul A, Kaosombatwattana U, Bhanthumkomol P, Bandidniyamanon W, Pausawasdi N, et al. The prevalence of steatohepatitis in chronic hepatitis B patients and its impact on disease severity and treatment response. Liver Int. 2017;37(4):542-51.
15. Choi HSJ, Brouwer WP, Zanjir WMR, de Man RA, Feld JJ, Hansen BE, et al. Nonalcoholic Steatohepatitis Is Associated With Liver-Related Outcomes and All-Cause Mortality in Chronic Hepatitis B. Hepatology. 2020;71(2):539-48.
16. Kim GA, Lim YS, An J, Lee D, Shim JH, Kim KM, et al. HBsAg seroclearance after nucleoside analogue therapy in patients with chronic hepatitis B: clinical outcomes and durability. Gut. 2014;63(8):1325-32.
17. Hui RWH, Seto WK, Cheung KS, Mak LY, Liu KSH, Fung J, et al. Inverse relationship between hepatic steatosis and hepatitis B viremia: Results of a large case-control study. J Viral Hepat. 2018;25(1):97-104.
18. Zhu L, Zhai X, Wang Q, Jiang J, Peng H, Song C, et al. Incidence and determinants of spontaneous hepatitis B surface antigen seroclearance and seroconversion in hepatitis B e antigen-negative chronic infection patients: A population-based prospective cohort. J Viral Hepat. 2018;25(12):1588-98.
19. Liu PT, Hwang AC, Chen JD. Combined effects of hepatitis B virus infection and elevated alanine aminotransferase levels on dyslipidemia. Metabolism. 2013;62(2):220-5.
20. Joo EJ, Chang Y, Yeom JS, Ryu S. Hepatitis B virus infection and decreased risk of nonalcoholic fatty liver disease: A cohort study. Hepatology. 2017;65(3):828-35.
21. Zhong GC, Wu YL, Hao FB, Rao XW, Yuan XW, Zhao Y, et al. Current but not past hepatitis B virus infection is associated with a decreased risk of nonalcoholic fatty liver disease in the Chinese population: A case-control study with propensity score analysis. J Viral Hepat. 2018;25(7):842-52.
22. Cheng YL, Wang YJ, Kao WY, Chen PH, Huo TI, Huang YH, et al. Inverse association between hepatitis B virus infection and fatty liver disease: a large-scale study in populations seeking for check-up. PLoS One. 2013;8(8):e72049.
23. Wang MM, Wang GS, Shen F, Chen GY, Pan Q, Fan JG. Hepatic steatosis is highly prevalent in hepatitis B patients and negatively associated with virological factors. Dig Dis Sci. 2014;59(10):2571-9.
24. Hu D, Wang H, Wang H, Wang Y, Wan X, Yan W, et al. Non-alcoholic hepatic steatosis attenuates hepatitis B virus replication in an HBV-immunocompetent mouse model. Hepatology International. 2018;12(5):438-46.
25. EASL-EASD-EASO Clinical Practice Guidelines for the management of non-alcoholic fatty liver disease. J Hepatol. 2016;64(6):1388-402.
26. Chalasani N, Younossi Z, Lavine JE, Diehl AM, Brunt EM, Cusi K, et al. The diagnosis and management of non-alcoholic fatty liver disease: practice Guideline by the American Association for the Study of Liver Diseases, American College of Gastroenterology, and the American Gastroenterological Association. Hepatology. 2012;55(6):2005-23.
27. Bozic M, Guzmán C, Benet M, Sánchez-Campos S, García-Monzón C, Gari E, et al. Hepatocyte vitamin D receptor regulates lipid metabolism and mediates experimental diet-induced steatosis. J Hepatol. 2016;65(4):748-57.
28. 2018 Practice Guidelines for the management of arterial hypertension of the European Society of Hypertension and the European Society of Cardiology: ESH/ESC Task Force for the Management of Arterial Hypertension: Erratum. J Hypertens. 2019;37(2):0000000000002026.
29. Eslam M, Newsome PN, Sarin SK, Anstee QM, Targher G, Romero-Gomez M, et al. A new definition for metabolic dysfunction-associated fatty liver disease: An international expert consensus statement. J Hepatol. 2020;73(1):202-9.
30. Cosentino F, Grant PJ, Aboyans V, Bailey CJ, Ceriello A, Delgado V, et al. 2019 ESC Guidelines on diabetes, pre-diabetes,

and cardiovascular diseases developed in collaboration with the EASD. Eur Heart J. 2020;41(2):255-323.
31. Mach F, Baigent C, Catapano AL, Koskinas KC, Casula M, Badimon L, et al. 2019 ESC/EAS Guidelines for the management of dyslipidaemias: lipid modification to reduce cardiovascular risk. Eur Heart J. 2020;41(1):111-88.
32. Chalasani N, Younossi Z, Lavine JE, Charlton M, Cusi K, Rinella M, et al. The diagnosis and management of nonalcoholic fatty liver disease: Practice guidance from the American Association for the Study of Liver Diseases. Hepatology. 2018;67(1):328-57.
33. Durrer Schutz D, Busetto L, Dicker D, Farpour-Lambert N, Pryke R, Toplak H, et al. European Practical and Patient-Centred Guidelines for Adult Obesity Management in Primary Care. Obes Facts. 2019;12(1):40-66.
34. Garcia-Tsao G, Lim JK. Management and treatment of patients with cirrhosis and portal hypertension: recommendations from the Department of Veterans Affairs Hepatitis C Resource Center Program and the National Hepatitis C Program. Am J Gastroenterol. 2009;104(7):1802-29.
35. Wiegand J, Berg T. The etiology, diagnosis and prevention of liver cirrhosis: part 1 of a series on liver cirrhosis. Dtsch Arztebl Int. 2013;110(6):85-91.
36. Yu MW, Shih WL, Lin CL, Liu CJ, Jian JW, Tsai KS, et al. Body-mass index and progression of hepatitis B: a population-based cohort study in men. J Clin Oncol. 2008;26(34):5576-82.
37. Wong GL, Chan HL, Yu Z, Chan AW, Choi PC, Chim AM, et al. Coincidental metabolic syndrome increases the risk of liver fibrosis progression in patients with chronic hepatitis B--a prospective cohort study with paired transient elastography examinations. Aliment Pharmacol Ther. 2014;39(8):883-93.
38. Chen CL, Yang HI, Yang WS, Liu CJ, Chen PJ, You SL, et al. Metabolic factors and risk of hepatocellular carcinoma by chronic hepatitis B/C infection: a follow-up study in Taiwan. Gastroenterology. 2008;135(1):111-21.
39. Xiong J, Zhang H, Wang Y, Wang A, Bian J, Huang H, et al. Hepatitis B virus infection and the risk of nonalcoholic fatty liver disease: a meta-analysis. Oncotarget. 2017;8(63):107295-302.

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Clinical approach to thyroid disease in adults

Athina-Lydia Mageiropoulou, Marina Michalaki

Abstract

The thyroid gland is an endocrine organ which secretes the thyroid hormones namely thyroxine and triiodothyronine which regulate vitally important functions of the human body, such as growth and metabolism. Thyroid diseases could be classified as those affecting its function (hyperthyroidism or hypothyroidism) or/and its morphology (goiter, thyroid nodules). The present review focuses on the clinical manifestations, evaluation, diagnosis, and therapy of thyroid diseases in adults.

Key words: *Thyroid; hypothyroidism; thyrotoxicosis; goiter; thyroid nodule; differentiated thyroid cancer*

INTRODUCTION

The thyroid gland is an endocrine organ, located in front of the trachea between the cricoid cartilage and the suprasternal notch, which secretes the thyroid hormones, namely thyroxine (T4) and triiodothyronine (T3). Thyroid hormones are iodinated tyrosine derivatives. The regulation of the synthesis and secretion of thyroid hormones is an example of negative feedback control by the hypothalamic thyrotropin-releasing hormone (TRH) and the pituitary thyrotropin (TSH) (Figure 1) [1]. Thyroid hormones are bound to plasma proteins, which are thyroxine-binding globulin (TBG), transthyretin and albumin and less than 1% of their total concentration remains free and active. Thyroid hormones act on their target tissues via TRa and TRβ nuclear receptor and promote brain and bone development in the fetus and during childhood, regulate the basic metabolic rate, appetite, heart rate, myocardial contraction, gastrointestinal motility and are involved in multiple functions of the human body [1]. Thyroid diseases could be classified as those affecting its function (hyperthyroidism or hypothyroidism) or/and its morphology (goiter, thyroid nodules). The diagnostic evaluation of thyroid diseases includes history, physical examination, laboratory measurements of thyroid

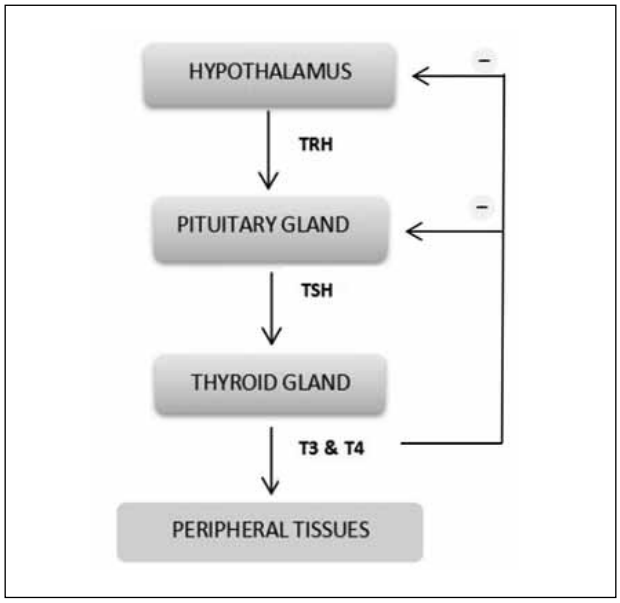

Figure 1. Hypothalamic-Pituitary-Thyroid axis, a paradigm of negative feedback regulation.

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hormones, TSH and antibodies against thyroid antigens and thyroglobulin in selected cases. Ultrasonography is very helpful in revealing thyroid nodules. Other imaging techniques like scintigraphy, computed tomography (CT), magnetic resonance imaging (MRI) or positron emission tomography (PET) could be indicated in certain circumstances.

Hypothyroidism

In hypothyroidism, the serum levels of T4 and T3 are low, due to impaired thyroid production (primary hypothyroidism) or very rarely due to hypophysis/hypothalamus defects (central or secondary hypothyroidism). The most common cause of primary hypothyroidism, in iodine sufficient areas, is the autoimmune Hashimoto's disease and iatrogenic causes such as thyroidectomy or radioiodine thyroid ablation [2]. The clinical manifestations of hypothyroidism are atypical, including weakness, feeling cold, difficulty concentrating, constipation, weight gain with poor appetite and menstrual disturbances in women of reproductive age. Hypothyroid patients, have dry skin, puffy face with edematous eyelids, bradycardia, and slow tendon reflex relaxation (most characteristic Achille). The diagnosis is made by measuring unbound T4 (free T4, FT4) and TSH, which in the case of primary hypothyroidism is high whereas in central hypothyroidism is inappropriately low (Table 1) [2] . The measurement of antibodies against thyroid peroxidase (Ab-TPO) is helpful for the diagnosis of Hashimoto's thyroiditis, and their levels are detectable in 90-100% of cases (Figure 2). The gold standard for the treatment of hypothyroidism of any cause, is the per os administration of levothyroxine (synthetic analog of thyroxine) pills in the morning on an empty stomach [3]. Approximately, 60-80% of ingested LT4 is absorbed in the jejunum and upper ileus. Absorption may be impaired in many circumstances, such as infection of *Helicobacter pylori*, atrophic gastritis, concurrent administration of certain drugs, as proton pump inhibitors, calcium and ferrum supplements and high fiber diet [4]. Recently, new forms of levothyroxine are available, like soft capsules or liquid preparations to overcome the difficulties with levothyroxine absorption [5]. The treatment goal is the replacement of the normal thyroid function which is evident when serum TSH levels are normalized in primary hypothyroidism or serum FT4 return to the upper reference range in central hypothyroidism [6]. There is a minority of hypothyroid patients, who do not feel well, despite adequate substitution of thyroid function with LT4. In those cases, the co-administration of T3 is an option [6].

Thyrotoxicosis

Thyrotoxicosis is the state of excessive circulating thyroid hormones, T3, T4 either due to their over-production and secretion from the thyroid gland, namely "hyperthyroidism" or due to their release from a destructed thyroid gland in case of thyroiditis or due to exogenous administration of high doses of thyroid hormones [7]. Extreme thyrotoxicosis leading to "thyroid storm" is a life-threatening situation [8]. The most common causes of hyperthyroidism are Grave's disease,

Table 1. *Laboratory evaluation of thyroid diseases.*

Disease	***Thyroid function tests***					
	T4	T3	TSH	Ab-TPO	Anti-Tg	TRab
Primary Hypothyroidism						
Subclinical	Normal	Normal	High	+ in HD	+ in HD	+ in HD
Overt	Low	Low	Very high*	+ in HD	+ in HD	+ in HD
Central Hypothyroidism						
	Low	Low	Normal/Low	-	-	-
Thyrotoxicosis						
Subclinical	Normal	Normal	Low	+ in GD	+ in GD	+ in GD
Overt	High	High	Undetectable	+ in GD	+ in GD	+ in GD

Abbreviations: TSH: Thyroid Stimulating Hormone; Ab-TPO: antibodies against thyroidal hyperoxidase; Anti-Tg: antibodies against thyroglobulin; TRabs: Thyrotropin Receptor antibodies; HD: Hashimoto's disease; GD: Graves 'disease; *Very high TSH: > 10mIU/L

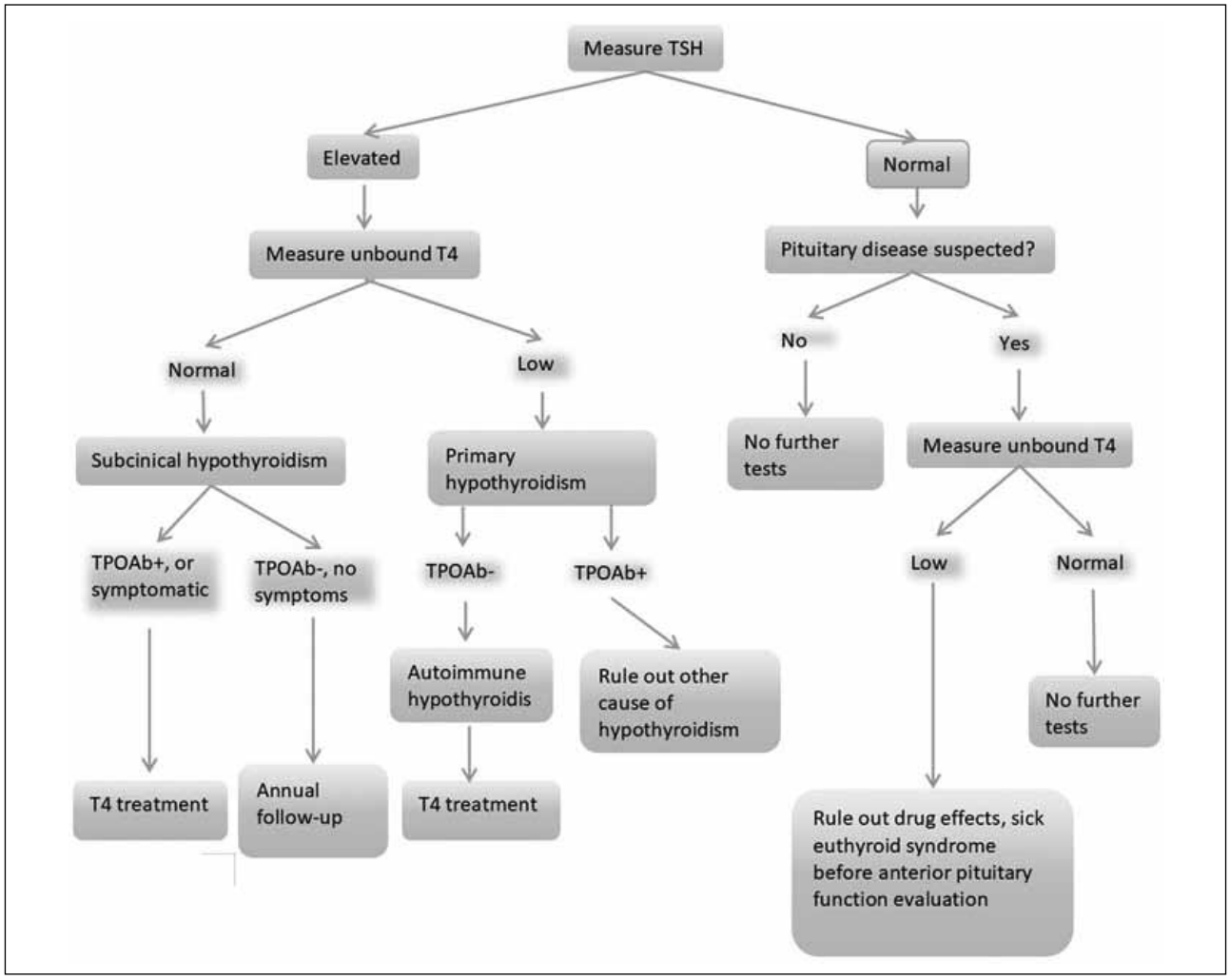

Figure 2. Diagnostic algorithm for evaluation of hypothyroidism.
Abbreviations: TSH: Thyroid Stimulating Hormone; Ab-TPO: antibodies against Thyroid peroxidase; T4: thyroxine.

toxic multinodular goiter and toxic adenomas [7]. The clinical manifestations of hyperthyroidism involve symptoms from multiple organs and are mostly the same regardless of the cause of thyrotoxicosis. They depend on the severity, duration, and patient's susceptibility to thyroid hormone excess [7]. Due to the enhanced metabolic rate, thyrotoxicosis can be presented as an unexplained weight loss despite an increased appetite, heat intolerance and easy sweating which makes their skin warm and moist. From the cardiovascular system, sinus tachycardia is the most common manifestation, occasionally associated with a sense of palpitations. As a result of a higher cardiac output, patients with a preexisting either known or subclinical heart failure or angina experience a deterioration of symptoms. Atrial fibrillation is common in patients >50 years of age and in about half of them it can remit after effective treatment of thyrotoxicosis. Other features are nervousness, hyperirritability, and insomnia, consequently leading to a sense of easy fatigability and impaired concentration. Also, diarrhea and mild steatorrhea, menstrual cycle disorders in women such as oligomenorrhea or amenorrhea, gynecomastia and impaired sexual function in men could be present. Osteopenia or even osteoporosis in the elderly, can occur due to long-standing elevated thyroid hormone levels, since they have a direct effect on bone resorption. In the elderly, fatigue and weight loss are predominant, whereas the other manifestations of thyrotoxicosis are missing, a syndrome known as "apathic thyreotoxicosis" which can be mistaken for depression.

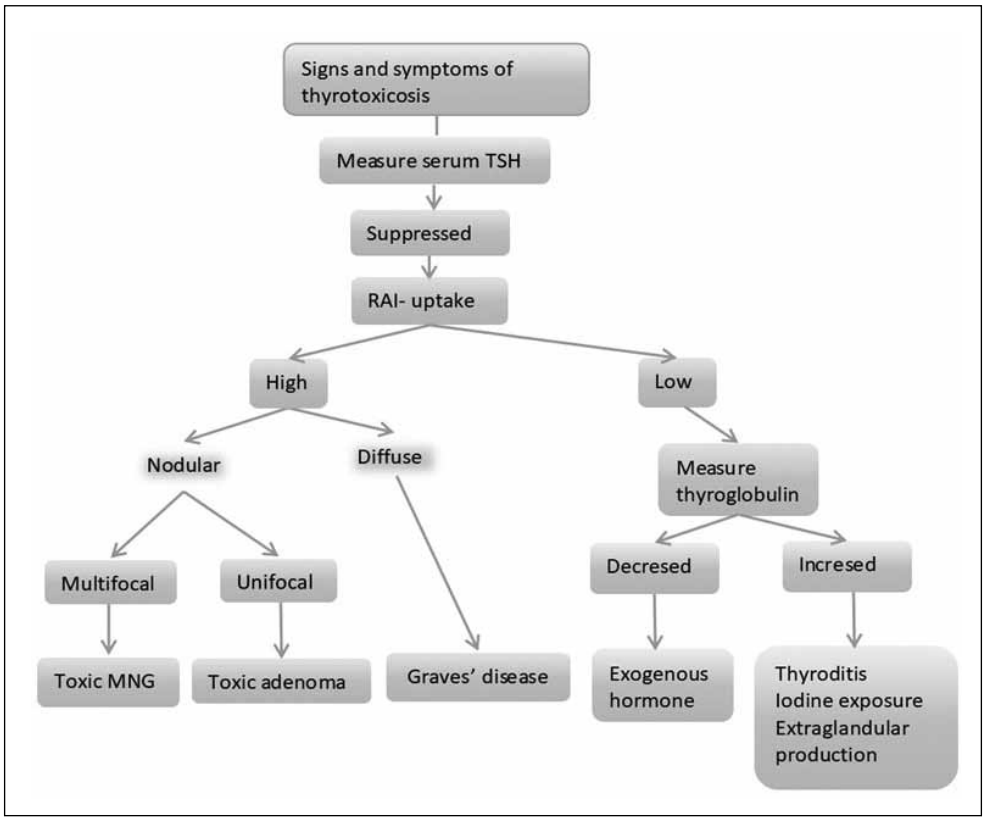

Figure 3. Diagnostic algorithm for evaluation of thyrotoxicosis.
Abbreviations: TSH: Thyroid Stimulating Hormone; RAI- ptake: Radioactive Iodine uptake; MNG: Multinodular Goiter.

The diagnosis of thyrotoxicosis is established by measuring high serum FT4, T3 and low or undetectable serum TSH levels (Table 1). In the extremely rare situation of a TSH secreting adenoma, serum TSH levels are inappropriately high [9] (Figure 3). In the case of Graves' disease, the measurement of thyroid stimulating immunoglobulin (TSI) can confirm the diagnosis [10].

Graves' disease (GD)

GD is an autoimmune disease and constitutes the most common cause of thyrotoxicosis. The disease is the result of the production of autoantibodies against the TSH receptor (TRAb), which mimic TSH action and cause hormonal hyperproduction and enlargement of the thyroid gland [11]. However, not all TRabs are stimulatory for the TSH receptor, but some are inhibitory. TRabs that stimulate the TSH receptor are also known as TSI. If TSIs predominate, then hyperthyroidism would be manifested in patients with GD. Moreover, it seems that the TSH receptor is expressed also in orbital and skin fibroblasts and orbital adipocytes thus GD can particularly be associated with Graves' orbitopathy in 30% of patients and very rarely with thyroid dermopathy [12]. The clinical manifestations of orbitopathy are exophthalmos, an anterior protrusion of the eyeball, motility disturbances manifesting with diplopia or pain, a sense of foreign body, orbital edema and conjunctival conditions, even visual loss if the optical nerve is compressed by the excessive tissue growth [13]. The course of Graves' disease has remissions and exacerbations. Thionamides (methimazole, carbimazole and propylthiouracil) inhibit the synthesis of thyroid hormones and are used for the treatment of GD. However, thionamides may have serious side-effects, although rare, like agranulocytosis and hepatotoxicity. Thus, more radical therapies are recommended when relapse occurs or drug therapy fails, such as, ablation with radioactive iodine or total thyroidectomy. The choice of treatment for GD depends on its activity and severity. In Graves' orbitopathy, control of risk factors, such as smoking and thyroid dysfunction, and local treatments are recommended in all patients. In active and severe Graves' orbitopathy, high doses of

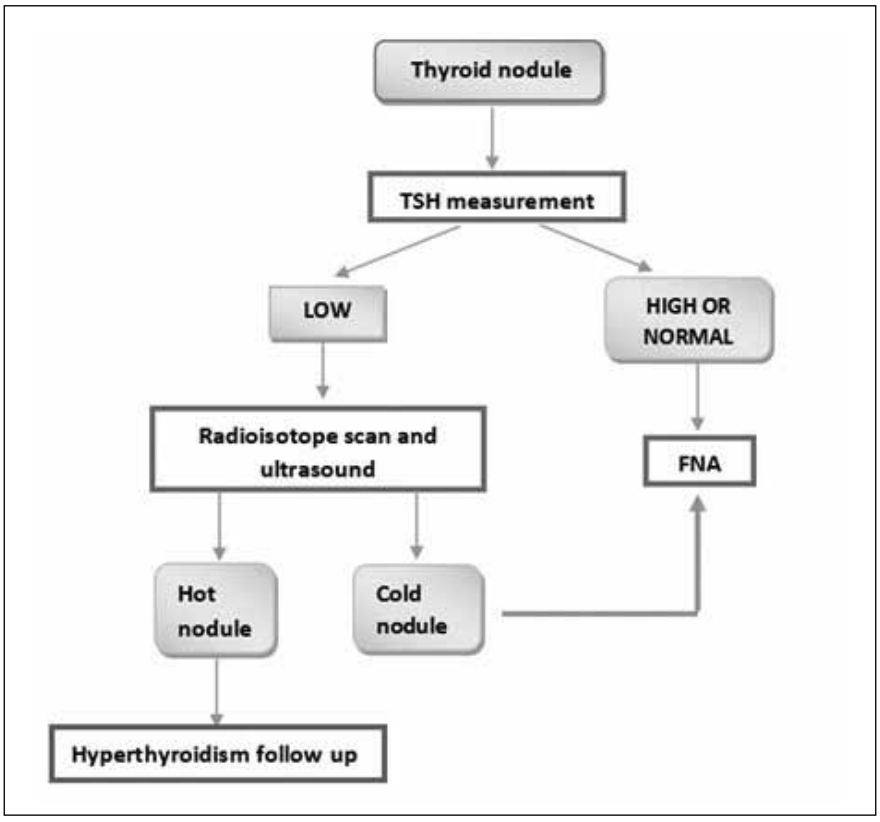

Figure 4. Clinical approach to thyroid nodule.
Abbreviations: TSH: Thyroid Stimulating Hormone; FNA: Fine Needle Aspiration

glucocorticoids or rituximab intravenously can be used. Recently, teprotumumab, an anti-IGF-1R monoclonal antibody, has exhibited very promising results [11, 13, 23].

Toxic multinodular goiter (TMNG) and toxic adenoma

Nodular thyroid disease commonly occurs in adults either as solitary or multiple nodules, which may be functional or not. Therefore, we can classify multinodular goiter in two categories, toxic multinodular goiter and non-toxic [14]. Toxic adenoma is a solitary functional thyroid nodule. Toxic multinodular goiter or toxic adenomas are characterized by functional autonomy caused mostly by activating mutations of the TSH receptor or the Gsa protein [14]. The clinical presentation of toxic MNG can be described as mild thyrotoxicosis. The treatment of toxic MNG is quite challenging [7]. Surgery is the definitive treatment of both nodular goiter and thyrotoxicosis. However, it is not the first treatment choice as the patients are usually elderly. Antithyroid drugs and beta blockers are usually used to control the symptoms of hyperthyroidism. Ablation with radioiodine could be a therapeutic option for elderly in whom thyroidectomy is contraindicated due to co-morbidities whereas it is the treatment of choice for toxic adenomas [7].

Thyroiditis

Thyroiditis is the inflammation and destruction of the thyroid gland and constitute a group of heterogenous diseases. The most common one is the *"subacute granulomatous thyroiditis or de Quervain's thyroiditis"* which is of uncertain origin [15,16]. A possible history of upper respiratory tract infection a few weeks prior to the onset of thyroiditis is usually reported. More frequently, it affects women of 30-50 years of age. The thyroid is painful and enlarged accompanied with fever and malaise. However, last year's many cases of painless subacute thyroiditis (SAT) have been reported, up to 6,25% in one series [16], thus the diagnosis is delayed. The course of thyroiditis follows 3 phases: a) a thyrotoxic phase due to the damage of follicular cells and the release of pre-formed thyroid hormones and thyroglobulin, b) a hypothyroid phase caused by

depletion of pre-formed thyroid hormones and the inability of the destructed gland to synthesize new and finally, c) the euthyroid phase during which the thyroid gland recovers, and its function is restored. Depending on the phase of illness, symptoms of thyrotoxicosis or hypothyroidism can occur. The diagnosis is confirmed by a high erythrocyte sedimentation rate (ESR) and low radioiodine uptake (<5%) in the face of thyrotoxicosis (T3 & T4 and TSH). White blood cell count is increased. Antibodies are negative. If there is a diagnostical doubt, FNA (fine needle aspiration) biopsy can be useful. Aspirin and NSAIDs (nonsteroidal anti-inflammatory drugs) could be used but in most cases high doses of glucocorticoids are indicated. The disease subsides in most cases, but a few patients may experience relapses and a prolonged course over many months. Permanent hypothyroidism occurs in about 15% of cases [17]. In addition, SAT has been described within two weeks of SARS-CoV-2 infection [16, 17].

"Acute thyroiditis" is rare and is caused by the suppurative infection of the thyroid, caused by gram-positive or gram-negative organisms [18,19]. It commonly occurs in children and young adults mostly when a piriform sinus, a remnant of the fourth branchial pouch that connects the oropharynx with the thyroid is present. Moreover, it is prevalent in immunosuppressed patients, such as those with the acquired immunodeficiency syndrome (AIDS) [18]. The patient exhibits acute thyroid pain and a tender goiter. Fever, dysphagia, and erythema over the thyroid are common, as are systemic symptoms of a febrile illness and lymphadenopathy. The erythrocyte sedimentation rate (ESR) and white cell count are usually increased, but the thyroid function usually is normal. FNA biopsy shows thyroid infiltration by polymorphonuclear leukocytes. Drainage is recommended and intravenous antibiotic therapy according to the analysis of the fluid obtained from the neck mass or of blood cultures [19].

Silent thyroiditis either postpartum or sporadic is painless, without fever and malaise and with a similar clinical course to subacute thyroiditis. This condition concerns 5% of women 3-6 months after delivery and has an autoimmune origin. A brief phase of mild thyrotoxicosis of 2-4 weeks is followed by a 4-12-week phase of hypothyroidism. Radioactive iodine uptake is suppressed whereas ESR is normal and there is a presence of TPO antibodies. The disease is mild and transient, and no treatment is needed. Beta-blockers can be used during the thyrotoxic phase to control adrenergic symptoms. An annual follow up is recommended thereafter because there is a possibility of the development of permanent hypothyroidism [18].

Several drugs can cause silent destructive thyroiditis, such as lithium, interferon alfa, interleukin-2, tyrosine kinase inhibitors, immune check point inhibitors and amiodarone.

Amiodarone's effects on the thyroid gland

Amiodarone is an antiarrhythmic drug type III, which influences the thyroid gland in many ways [20]. The chemical structure of amiodarone is related to thyroid hormones and contains two iodine atoms. Amiodarone metabolism in the liver releases approximately 3 mg of inorganic iodine into the systemic circulation per 100 mg of amiodarone ingested, meaning that the administration of a regular dose of 200-400 mg of amiodarone per day loads the body with pharmacological doses of iodine. Furthermore, amiodarone alters thyroid hormone metabolism, may be toxic for the thyroid gland and is stored in adipose tissue and for this reason high levels of plasma iodine can persist even for more than 6 months following drug withdrawal. Normally, the first few weeks after the administration of amiodarone a transient slight elevation of serum TSH and FT4 occurs while T3 drops. However, when an underlying thyroid disease exists, amiodarone can cause either thyrotoxicosis or hypothyroidism. There are, two different types of thyrotoxicosis (AIT - amiodarone induced thyrotoxicosis); the first one is related to already existing thyroid disorder (Graves' disease or multinodular goiter), in which the synthesis of thyroid hormones becomes excessive as a result of increased exposure to iodine (Jod-Basedow phenomenon); the second one appears in patients without any thyroid disorder and is a result of a drug induced lysosomal activation which causes destructive thyroiditis. The differential diagnosis between the two types is challenging and crucial because treatment completely differs between the two types of thyrotoxicosis [20]. The diagnosis is based on history of any thyroid disease, colored doppler and iodine uptake on scintigraphy, because unlike type 2, type 1 is characterized by increased vascularity and iodine uptake. The treatment of type 1 is antithyroid drugs whereas in type 2 high doses of glucocorticoids are given. Frequently, both treatments are used because the differential diagnosis between the two types is uncertain or mixed types exist. Rarely, drug therapies fail, and total thyroidectomy is indicated. The withdrawal of amiodarone is a matter of debate but in most cases is desirable. Additionally, the

decision to withdraw is not urgent since the half-life of amiodarone is 100 days [20].

Non-thyroidal illness syndrome

An interesting phenomenon found in patients with an acute severe non-thyroidal disease or fasting is the alterations in thyroid function tests namely "non-thyroidal illness syndrome" or "euthyroid sick syndrome" [21, 22]. The syndrome does not have any deleterious consequences in these patients, so it does not need treatment, and it seems that it is an adaptation of the organism to inhibit catabolism and save energy. The pathogenesis is not clearly described yet, but inflammatory cytokines such as IL-6, and drugs are involved. The most common pattern in the non-thyroidal illness syndrome is a decrease in total and free levels of T3 with normal levels of T4 and TSH. The severity of illness can correlate to the magnitude of fall of T3 levels. Peripheral deiodination of T4 to T3 is impaired leading to an increase of (inactive) reverse T3 (rT3), due to the inactivation of DI and DII deiodinase isoenzymes. In very severe disease or critical illness, patients may exhibit low T4 and TSH levels, associated with poor prognosis [22].

Goiter, thyroid nodular disease and thyroid carcinoma

Goiter is the enlargement of the thyroid gland due to the irregular growth of follicular cells [14]. Iodine deficiency, autoimmune disease or dyshormonogenesis cause hypothyroidism which in turn increases TSH secretion to restore normal thyroid hormone levels but concurrently TSH has trophic effects on the follicular cells. However, in most of the goiters no cause is identified. Long-standing goitrogenesis gives rise to the development of focal or nodular hyperplasia [14]. Nodular thyroid disease commonly occurs among adults either as solitary or multiple nodules, which may be functional or not, as mentioned above.

Thyroid nodules are common radiological findings, depicted as a discrete lesion in the thyroid gland that is radiologically distinct from the surrounding parenchyma [23]. The major clinical concerns related to nodules are the exclusion of malignancy (4-6,5% of thyroid nodules), evaluation of functional status and assessment for the presence of pressure symptoms [23]. Non-functioning benign nodules could be a colloids, Hashimoto's thyroiditis, cysts, and more rarely follicular adenomas [24]. Papillary thyroid cancer is the most common type of thyroid cancer representing approximately 95% of all cases [25]. It originates from the epithelial follicular cells, is well differentiated and in most cases mortality is nearly zero. Other types of thyroid cancer are follicular and Hurthle Cell (oncocytic) carcinomas also derived from follicular cells with less favorable prognosis (10 years mortality around 10%) [25, 26]. Approximately 1-2% of thyroid cancers are medullary carcinomas, which originate from c-cells thus producing calcitonin which serves as a marker. It can be either sporadic or familial (MEN 2A, MEN 2B, familial MTC with other features) in 25% of cases and thus all patients diagnosed with medullary thyroid cancer (MTC) should be tested for RET (*Ret* Proto-Oncogene) mutations, pheochromocytoma, and hyperparathyroidism [25]. MTC has less favorable prognosis depending on the stage of the disease with mortality rate ranging from 0% to 100% from stage I to stage IV [27]. Finally, anaplastic carcinomas are rare (<1%), poorly differentiated, very aggressive with a poor prognosis. They spread locally into surrounding tissues like trachea, larynx, and laryngeal nerves. Also, thyroid lymphomas or metastases from other primary tumors such as renal carcinomas and melanomas could be detected [25].

During the initial evaluation a detailed history assessing risk factors, such as: head or neck radiation during childhood, total body irradiation for bone marrow transplantation, exposure to ionizing radiation in young age, family history of papillary or medullary carcinoma or thyroid cancer syndromes, presence of rapidly enlarging nodule or fixed nodule to surrounding tissues and pressure symptoms such as vocal cord paralysis and hoarseness is required [23].

Laboratory assessment should begin with the measurement of TSH levels. Low serum TSH suggests an overt or subclinical hyperthyroidism (Figure 4). A radionuclide thyroid scan should be performed next to determine the functional status of the nodule. Nodules may appear 'hot', 'warm' or 'cold' depending on whether they absorb radioisotopes more, less than or equally to the remaining normal thyroid tissue. Only non-functioning/cold nodules have a malignancy possibility, and these nodules should be punctuated to obtain specimens for cytological examination (FNA) [26]. The indication for the FNA of a cold thyroid nodule depends on its size and sonographic features (hypoechogenicity, irregular margins, shape with vertical bigger than transverse dimension, microcalcifications are indicative of malignancy). The cytological diagnosis is mostly reported according to the "2017 Bethesda System" [24]. There

are six diagnostic categories and each one predicts the risk of malignancy: (i) nondiagnostic or unsatisfactory (5-10%.); (ii) benign (1-3%); (iii) atypia of undetermined significance (AUS) or follicular lesion of undetermined significance (FLUS) (15%); (iv) follicular neoplasm or suspicious for a follicular neoplasm (10-40%); (v) suspicious for malignancy (50-75%); and (vi) malignant (99%). The clinical management of non-functioning thyroid nodules depends on the cytopathological findings and the implying risk of malignancy. Thus, we do not operate on benign nodules which do not pressure adjacent vital structures, but we periodically evaluate them with ultrasound. For the unspecified categories (iii and IV), there are several options: where available, mutational analysis should be performed or repeat FNA after 6 months or lobectomy. Patients with cytological findings suggesting thyroid carcinoma should be referred for surgery. The gold standard for the treatment of differentiated thyroid carcinoma (DTC), which accounts for approximately of 90% of thyroid cancer, is total thyroidectomy followed by radioiodine therapy [26]. More recently, it became clear that the mortality rate is not parallel to the morbidity for DTC. New staging systems have been developed to predict disease persistence or recurrence which is independent of death. For example, DTC patients with small infiltrated cervical lymph had 10year survival nearly 100%, especially if they were younger than 55 years old. Thus, the American Thyroid Association (ATA) in 2015 categorized DTC patients as low, intermediate, and high risk for recurrence based on their clinicopathological characteristics. For low-risk patients, alternative therapeutical options are lobectomy instead of total thyroidectomy, avoidance of radioiodine therapy and even active surveillance without surgery for micropapillary (<1cm) thyroid carcinomas especially for elderlies with several co-morbidities [28]. In contrast, for high-risk patients with advanced DTC or MTC or anaplastic carcinomas besides total thyroidectomy, tyrosine kinase inhibitors can be used and seem very promising [27,29].

Conclusions and Future directions

Thyroidal diseases are complex including either functional or morphological or both disorders of the hypothalamic-pituitary-thyroid axis. The differential diagnosis in most cases is difficult and demands a thorough history and physical examination, targeted laboratory tests, ultrasonographic examination of the gland and in selected cases scintigraphy, CT, MRI or PET scans. Most of thyroidal diseases are treated successfully, however there are still unmet therapeutic needs especially for patients with Graves' orbitopathy and advanced thyroid cancer. Furthermore, issues remain regarding the quality of life of patients even with mild thyroid diseases such as hypothyroidism. Novel diagnostic tools and treatments are expected in the near future.

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REFERENCES

1. Stathatos N. Thyroid Physiology. Med Clin North Am. 2012;96(2):165-73.
2. Garber J, Cobin R, Gharib H, Hennessey J, Klein I, Mechanick J, et al. Clinical Practice Guidelines for Hypothyroidism in Adults: Cosponsored by the American Association of Clinical Endocrinologists and the American Thyroid Association. Endocr Pract. 2012;18(6):988–1028.
3. Pearce S, Brabant G, Duntas L, Monzani F, Peeters R, Razvi S et al. 2013 ETA-Guideline: Management of Subclinical Hypothyroidism. Europea Thyroid J. 2013;2(4):215-28.
4. Virili C, Antonelli A, Santaguida M, Benvenga S, Centanni M. Gastrointestinal Malabsorption of Thyroxine. Endocr Rev. 2018;40(1):118-36.
5. Nagy E, Perros P, Papini E, Katko M, Hegedüs L. New Formulations of Levothyroxine in the Treatment of Hypothyroidism: Trick or Treat? Thyroid. 2021;31(2):193-201.
6. Jonklaas J, Bianco A, Cappola A, Celi F, Fliers E, Heuer H et al. Evidence-Based Use of Levothyroxine/Liothyronine Combinations in Treating Hypothyroidism: A Consensus Document. Eur Thyroid J. 2021;10(1):10-38.
7. Ross D, Burch H, Cooper D, Greenlee M, Laurberg P, Maia A et al. 2016 American Thyroid Association Guidelines for Diagnosis and Management of Hyperthyroidism and Other Causes of Thyrotoxicosis. Thyroid. 2016; 26(10):1343-421.
8. Ishii M. Endocrine Emergencies with Neurologic Manifestations. Continuum (Minneap Minn). 2017;23(3):778-801.
9. Beck-Peccoz P, Brucker-Davis F, Persani L, Smallridge R, Weintraub B. Thyrotropin-Secreting Pituitary Tumors. Endocr Rev. 1996; 17(6):610-38.
10. Kahaly G. Management of Graves Thyroidal and Extrathyroidal Disease: An Update. J Clin Endocrinol Metab. 2020;105(12):3704-20.
11. Smith T, Hegedüs L. Graves' Disease. N Engl J Med. 2016;375(16):1552-65.

12. Taylor P, Zhang L, Lee R, Muller I, Ezra D, Dayan C, et al. 2019. New insights into the pathogenesis and nonsurgical management of Graves orbitopathy. Nature Reviews Endocrinology, 16(2), pp.104-16.
13. Bartalena L, Kahaly G, Baldeschi L, Dayan C, Eckstein A, Marcocci C et al. The 2021 European Group on Graves' orbitopathy (EUGOGO) clinical practice guidelines for the medical management of Graves' orbitopathy. Eur J Endocrinol. 2021;185(4):G43-67.
14. Krohn K, Führer D, Bayer Y, Eszlinger M, Brauer V, Neumann S, et al. Molecular Pathogenesis of Euthyroid and Toxic Multinodular Goiter. Endocr Rev. 2004;26(4):504-24.
15. Stasiak M, Lewiński A. New aspects in the pathogenesis and management of subacute thyroiditis. Rev Endocr Metab Disord. 2021; 22(4):1027-39.
16. Stasiak M, Michalak R, Stasiak B, Lewinski A. Clinical characteristics of subacute thyroiditis is different than it used to be - current state based on 15 years own material. Neuro Endocrinol Lett. 2019;39(7):489–95.
17. Pal R, Banerjee M. COVID-19 and the endocrine system: exploring the unexplored. J Endocrinol Invest 2020; 43(7):1027-31.
18. Pearce E, Farwell A, Braverman L. Thyroiditis. N Engl J Med. 2003;348(26):2646-55.
19. AU Paes JE, Burman KD, Cohen J, Franklyn J, McHenry CR, Shoham S, et al. Acute bacterial suppurative thyroiditis: a clinical review and expert opinion. Thyroid. 2010;20(3):247-55.
20. Basaria S, Cooper D. Amiodarone and the thyroid. Am J Med. 2005;118(7):706-14.
21. Chopra I. Euthyroid Sick Syndrome: Is It a Misnomer? J Clin Endocrinol Metab. 1997;82(2):329-34.
22. Fliers E, Bianco A, Langouche L, Boelen A. Thyroid function in critically ill patients. Lancet Diabetes Endocrinol. 2015;3(10):816-25.
23. Hegedüs L. Clinical practice. The Thyroid Nodule. N Engl J Med. 2004;351(17):1764-71.
24. Cibas E, Ali S. The 2017 Bethesda System for Reporting Thyroid Cytopathology. Thyroid. 2017;27(11):1341-6.
25. Fagin J, Wells S. Biologic and Clinical Perspectives on Thyroid Cancer. N Engl J Med. 2016; 375(11):1054-67.
26. Haugen B, Alexander E, Bible K, Doherty G, Mandel S, Nikiforov Y et al. 2015 American Thyroid Association Management Guidelines for Adult Patients with Thyroid Nodules and Differentiated Thyroid Cancer: The American Thyroid Association Guidelines Task Force on Thyroid Nodules and Differentiated Thyroid Cancer. Thyroid. 2016;26(1):1-133.
27. Wells S, Asa S, Dralle H, Elisei R, Evans D, Gagel R et al. Revised American Thyroid Association Guidelines for the Management of Medullary Thyroid Carcinoma. Thyroid. 2015;25(6):567-610.
28. Chou R, Dana T, Haymart M, Leung A, Tufano R, Sosa J et al. Active Surveillance Versus Thyroid Surgery for Differentiated Thyroid Cancer: A Systematic Review. Thyroid. 2022;32(4):351-67.
29. Fugazzola L, Elisei R, Fuhrer D, Jarzab B, Leboulleux S, Newbold K et al. 2019 European Thyroid Association Guidelines for the Treatment and Follow-Up of Advanced Radioiodine-Refractory Thyroid Cancer. Eur Thyroid J. 2019;8(5):227-45.

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Chronic Obstructive Pulmonary Disease Treatment Guidelines

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Abstract

Chronic obstructive pulmonary disease (COPD) affects one-tenth of the world's population and has been identified as a major global unmet health need by the World Health Organization. Increased healthcare resource use is common among patients with frequent exacerbations, and exacerbations are a major cause of the high 30-day hospital re-admission rates associated with COPD. Timely and appropriate maintenance pharmacotherapy, particularly dual bronchodilators for maximizing bronchodilation, can significantly reduce exacerbations in patients with COPD. Additionally, multidisciplinary disease-management programs include pulmonary rehabilitation, follow-up appointments, aftercare, inhaler training, and patient education that can reduce hospitalizations and readmissions for patients with COPD. With the availability of newer pharmacotherapy options, treatment recommendations are made on the basis of a review of the latest literature and directed by symptom burden and health care utilization.

Key words: *COPD; GOLD; bronchodilators; exacerbations; inhaler corticosteroids; LAMA/LABA*

INTRODUCTION

Chronic Obstructive Pulmonary Disease (COPD) is a major social, economic and health burden since it is the third most common cause of death worldwide [1]. COPD prevalence in Europe ranges between 4% and 10% [2]. COPD is underdiagnosed and often misdiagnosed, which contributes to the continuing increases in the prevalence, morbidity, and mortality. It is a common, preventable, and treatable disease which is characterized by persistent respiratory symptoms and airflow obstruction that is due to airway and/or alveolar abnormalities. These are usually caused by significant exposure to noxious particles and/or gases. It has been repeatedly suggested that management of the very large number of patients with COPD can be improved by the development and implementation of evidence-based diagnostic/screening and treatment guidelines. The objective of this review is to overhaul the evidence recently published in order to define COPD characteristics able to suggest a therapeutic algorithm.

BASIC MECHANISMS OF COPD

COPD is characterized by a partially reversible airflow obstruction and an abnormal inflammatory response in the lungs. The latter represents the innate and adaptive immune responses to long-term exposure to noxious particles and gases, particularly cigarette smoke. All cigarette smokers have some inflammatory burden in their lungs, but those who develop COPD have an abnormal response to inhaling toxic agents. This amplified response may result in mucous hypersecretion (chronic bronchitis), tissue destruction (emphysema), and disruption of normal repair and defense mechanisms, causing

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small airway inflammation and fibrosis (bronchiolitis). These pathological changes result in increased resistance to airflow in the small airways, increased compliance of the lungs, air trapping, and progressive airflow obstruction-all of which are characteristic features of COPD. Inflammation is present in the lungs, particularly the small airways, of all people who smoke. This normal protective response to the inhaled toxins is amplified in COPD, leading to tissue destruction, impairment of the defense mechanisms that limit such destruction, and disruption of the repair mechanisms. In general, the inflammatory and structural changes in the airways increase with disease severity and persist even after smoking cessation. Besides inflammation, two other processes are involved in the pathogenesis of COPD-an imbalance between proteases and antiproteases and an imbalance between oxidants and antioxidants in the lungs [3]. Neutrophils, macrophages and T lymphocytes are inflammatory cells which release a plethora of cytokines and mediators that participate in the disease process, such as Leukotriene B4, TNFa, TGFβ, IL-1B and IL-6. Increased production of proteases (cathepsins G, E, A, L, metalloproteases, protease 3 and serine proteases elastase) and inactivation of antiproteases including a1-antitrypsin and secretory leucoprotease inhibitor results in imbalance. Oxidative stress can lead to inactivation of antiproteases or stimulation of mucous production. It can also amplify inflammation by enhancing transcription factor activation (such as nuclear factor κB) and hence gene expression of pro-inflammatory mediators [3].

AETIOLOGY OF COPD

Worldwide, the most common cause of COPD is tobacco smoking. Cigarette smokers have a higher prevalence of respiratory symptoms and lung function abnormalities, a greater decline in FEV1 and a greater mortality rate than non-smokers. There is a significant correlation between indoor and outdoor air pollution including burning of the wood and other biomass fuels and the incidence of COPD. Genetic factors such as severe hereditary deficiency of alpha-1 antitrypsin, the gene encoding matrix metalloproteinase 12 (MMP-12) and glutathione S-transferase, as well as aging and female sex have been related to a decline in lung function or risk of COPD. Chronic bronchitis, any type of infections, asthma and airway hyperreactivity may increase the frequency and severity of exacerbations [4].

DIAGNOSTIC CRITERIA AND CLASSIFICATION OF SEVERITY OF COPD

Early screening for COPD is based on early detection in primary care medicine. Recent studies have highlighted the need for the application of scores to increase diagnosis of COPD by using spirometers in general primary practice [5]. The objective of screening is to accurately detect airflow obstruction, even in patients with few symptoms. COPD should be considerate in any patient who has dyspnea, chronic cough and sputum production and/or a history of exposure in risk factors. Spirometry is the most objective method to make the diagnosis. The presence of post-bronchodilator FEV1/FVC <0.70 confirms the airflow limitation and thus of COPD in patients with appropriate symptoms and significant exposures to noxious stimuli. Despite its good sensitivity, peak expiratory flow measurement alone cannot be reliably used as the only diagnostic test because of its weak specificity [6].

There is a wide use of the FEV1/FVC ratio as the primary factor in determining the presence or absence of airway obstruction but there are differences of opinion about what value of FEV1/FVC should be used for this purpose. Currently, there are two main schools of thought; those that advocate the use of the GOLD fixed 70% ratio and those that instead advocate the use of the lower limit of normal (LLN) for the FEV1/FVC ratio. There is some evidence that individuals with an FEV1/FVC ratio below 70% tend to have more significant lung disease and a higher mortality. Numerous studies, however, have shown that the GOLD threshold overestimates airway obstruction in the elderly and the tall and underestimates it in the young and the short. The main predictors beyond the FEV1/FVC ratio for an expert diagnosis of COPD were the FEV1 % predicted, and the residual volume/total lung capacity ratio (RV/TLC). Adding FEV1 and RV/TLC to GOLD or LLN improved the diagnostic accuracy, resulting in a significant reduction of up to 50% of the number of misdiagnoses. GOLD criteria over-diagnose COPD, while LLN definitions under-diagnose COPD in elderly patients as compared to an expert panel diagnosis. Incorporating FEV1 and RV/TLC into the GOLD-COPD or LLN-based definition brings both definitions closer to expert panel diagnosis of COPD, and to daily clinical practice [7,8].

The Global Initiative for Chronic Obstructive Lung Disease (GOLD) proposes that patients are stratified according to disease severity, with the incorporation of symptoms determined with the modified Medical Re-

search Council (mMRC) scale or the state of health using the COPD Assessment Test (CAT), as well as the patient's history of exacerbations and post-bronchodilator (pb) FEV1%. Patients are classified according to risk: low risk (pbFEV1%≥50% or <2 exacerbations in the previous year) and high risk (pbFEV1%<50% or ≥2 exacerbations in the previous year). The risk index must be determined according to airflow limitation and history of exacerbations. Depending on the symptomatic impact, patients are classified as having less symptoms (CAT<10 or mMRC 0–1) or more symptoms (CAT≥10 or mMRC≥2). Thus, four categories are identified: A (low risk, less symptoms), B (low risk, more symptoms), C (high risk, less symptoms), D (high risk, more symptoms). The proposed therapeutic approach is different for each group [9,10] (Figure 1).

COPD MANAGEMENT

Reduction in the risk of exacerbation, along with symptom management, is the cornerstone of the current strategy for management of COPD. The main components of COPD management are appropriate pharmacotherapy (that addresses both symptom management and exacerbation prevention), promotion of smoking cessation, pulmonary rehabilitation, and regular follow-up monitoring for disease progression.

Smoking cessation and risk factor avoidance

Smoking cessation remains the only intervention definitively known to halt the progression of COPD, and several studies have demonstrated that early smoking cessation has the potential to slow down or even reverse accelerated decline in lung function, highlighting the importance of intervention in early disease [11]. As previously shown by Anthonisen et al., special programs in supporting smoking cessation can achieve a reduction in terms of all-cause mortality, even if those interventions are successful only in a minority of patients [12]. One explanation of a better survival in former smokers is partly attributable to the prevention of smoking damage over time -lower functional decay of the lung [13] and increased risk of cancer and cardiovascular diseases in smokers [12] and partly to the greater pharmacological efficacy of compounds containing ICS [14]. Interestingly, in a recent Delphi consensus project run in Italy, the most effective step to reduce lung functional decline were considered by the 207 specialists interviewed to be smoking cessation [15].

Pharmacotherapy and nicotine replacement reliably increase long-term smoking abstinence rates [16]. Legislative smoking bans and counseling, delivered by health-care professionals, improve quit rates. Nonethe-

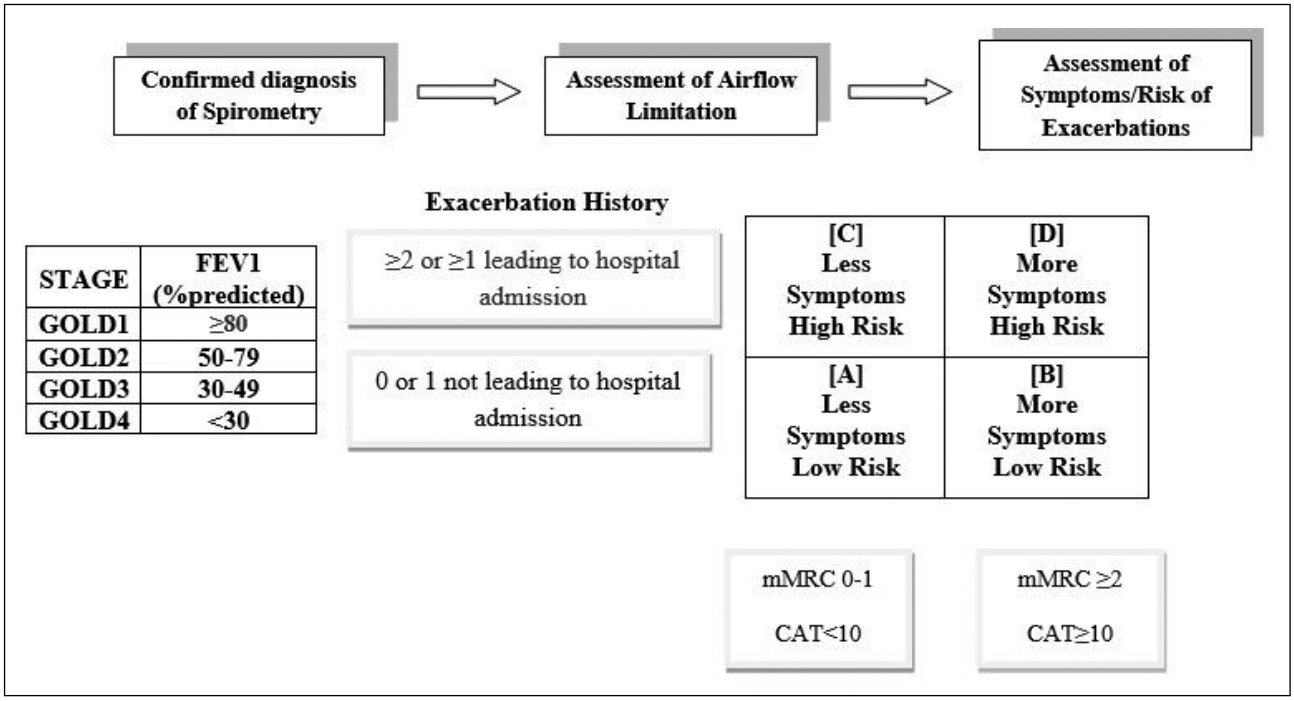

Figure 1. ABCD assessment tool.

less, the effectiveness and safety of e-cigarettes as a smoking cessation aid is uncertain at present [17].

Vaccination for stable COPD

Patients with COPD and other chronic respiratory diseases are especially vulnerable to viral and bacterial pulmonary infections, which are major causes of exacerbations, hospitalization, disease progression, and mortality in COPD patients [18]. The WHO and CDC recommend SARS-CoV-2 and influenza vaccination as they reduce serious illness in COPD patients [19]. While COPD itself is not a risk factor for acquiring a SARS-CoV-2 infection, existing lung damage due to COPD means people are more likely to experience severe complications of COVID-19. Recent studies highlight that having COPD can increase a person's risk of hospitalization, ICU admission, and death from COVID-19. The 23-valent pneumococcal polysaccharide vaccine (PPSV23) has shown lower incidence of community-acquired pneumonia aged <65 years with an FEV1<40% predicted, while the 13-valent conjugate pneumococcal vaccine (PCV13) in the group of patients >65 years reduced bacteremia and serious invasive pneumococcal disease [20]. Furthermore, the CDC recommends Tdap (dTaP/dTPa) vaccination to protect against pertussis for adults with COPD who were not vaccinated in adolescence and Zoster vaccine to protect against shingles for adults with COPD aged >50 years [21].

Oxygen Therapy and Ventilatory Support

The long- term administration of oxygen (>15 hours per day) to patients with chronic respiratory failure has shown to increase survival in patients with severe resting hypoxemia [22]. In patients with stable COPD and moderate resting or exercise-induced arterial desaturation, prescription of long-term oxygen does not lengthen time to death or first hospitalization or provide sustained benefit in health status, lung function and 6-minute walk distance [23,24].

The role of NIV in COPD is to decrease work of breathing and improve respiratory mechanics through effects on several pathophysiologic abnormalities present in severe COPD. Hyperinflation together with other pathobiological mechanisms related to muscle dysfunction in severe COPD lead to diaphragm muscle atrophy. The combination of diaphragm muscle atrophy and the airflow obstruction central to COPD pathophysiology leads to increased respiratory muscle load. The goal of NIV in COPD is to offset this diaphragmatic dysfunction and achieve control of spontaneous breathing with near-abolition of diaphragm activity, reducing chronic hypercapnia. Although the direct impact that impaired gas exchange has on work of breathing is unclear, there is evidence that hypoxemia can impact skeletal muscle strength and endurance and that chronic hypercapnia can induce skeletal muscle dysfunction. In addition, emerging data indicate that chronic hypercapnia suppresses innate immunity and that a reduction in CO_2 levels may have a mechanistic effect in reduction of COPD exacerbations leading to hospital admissions [25]. So, noninvasive ventilation (NIV) in the form of noninvasive positive pressure ventilation (NPPV) is the standard of care for decreasing morbidity and mortality in patients hospitalized with an exacerbation of COPD and acute respiratory failure, particularly in those with pronounced daytime persistent hypercapnia PCO2 >52mmHg [26, 27]. In patients with both COPD and obstructive sleep apnea there are clear benefits associated with the use of continuous positive airway pressure (CPAP) to improve both survival and the risk of hospital admissions [28].

Pulmonary Rehabilitation

Pulmonary rehabilitation is a comprehensive intervention based on a thorough patient assessment followed by patient-tailored therapies, which include, but are not limited to, exercise training, education and behavior change, designed to improve the physical and emotional condition of people with chronic respiratory disease and to promote the long-term adherence of health-enhancing behaviors [29].

Physical inactivity is an important determinant of health-related quality of life in patients with COPD and is a predictor of hospitalization and mortality [30]. Physical deconditioning that follows leads to even more exertional dyspnea and further deconditioning. Studies have demonstrated reduced skeletal muscle strength even in patients with mild airflow obstruction as well as in smokers without airflow obstruction, suggesting that physical deconditioning is present in early disease [31,32].

Traditionally, pulmonary rehabilitation was prescribed for patients with severe disease. However, recent data, including a systematic review of the available data for pulmonary rehabilitation in patients with mild COPD, showed evidence of improved exercise capacity and health-related quality of life, and improvement in 6-min walk test, suggesting their potential benefit even in early disease [31].

There is currently not enough capacity to deliver conventional pulmonary rehabilitation for large numbers of patients with early disease, and new modes of increasing exercise and fitness levels such as digital interventions will need to be tested in the context of these patient groups [33].

Totally, pulmonary rehabilitation not only improves dyspnea, health status and exercise tolerance in patients with stable COPD, but also, leads to a reduction in symptoms of anxiety and depression and reduces hospitalization among those with a recent exacerbation (<4 weeks from prior hospitalization).

Surgical Interventions

Lung Volume Reduction Surgery (LVRS) is a surgical technique that may be beneficial for some patients with advanced emphysema who have poor control of their disease despite maximal medical therapy. LVRS entails reducing the lung volume by wedge excision of emphysematous tissue. Subsequent modifications to LVRS include non resectional lung volume reduction [34,35]. LVRS reduces the elastic recoil pressure of the lung and thus improves expiratory flow rates and reduces exacerbations [36,37].

Bullectomy is the surgical removal of a bulla, which is a dilated air space in the lung parenchyma measuring more than 1 cm. It is carried out in selected patients and it is associated with decreased dyspnea, improved lung function and exercise tolerance [38].

Due to the morbidity and mortality associated with LVRS, less invasive bronchoscopic approaches to lung reduction have been examined. These include a variety of bronchoscopic procedures such as endobronchial valves, lung coils and vapor ablation. Although these techniques differ markedly from one another, they are similar in their objective to decrease thoracic volume to improve lung, chest wall and respiratory muscle mechanics [39].

Last but not least, lung transplantation in appropriately selected patients with severe COPD has been shown to improve quality of life and functional capacity, but not prolong survival [40].

PHARMACOLOGICAL THERAPY FOR COPD

Bronchodilators

Bronchodilators are central to the treatment of COPD, notwithstanding that there is often limited reversibility of airflow obstruction. The existing drug classes ($beta_2$-agonists and muscarinic receptor antagonists) work by relaxing airway smooth muscle tone, leading to reduced respiratory muscle activity and improvements in ventilatory mechanics, making it easier for patients to breathe. Bronchodilation aims at alleviating bronchial obstruction and airflow limitation, reducing hyperinflation, and improving emptying of the lung and exercise performance [41,42].

The principal action of $beta_2$-agonists is to relax airway smooth muscle by stimulating $beta_2$-adrenergic receptors, which increases cyclic AMP and produces functional antagonism to bronchoconstriction. There are short-acting (SABA) and long-acting (LABA) $beta_2$-agonists. Regular and as-needed use of SABAs improves FEV_1 and symptoms. LABAs show duration of action of 12 or more hours and do not preclude additional benefit from as-needed SABA therapy [43]. Formoterol and salmeterol are twice-daily LABAs that significantly improve FEV_1 and lung volumes, dyspnea, exacerbation rate and number of hospitalizations, but have no effect on mortality rate or decline of lung function.

Additionally, antimuscarinic drugs block the bronchoconstrictor effects of acetylcholine on M3 muscarinic receptors expressed in airway smooth muscle [44]. Short-acting (SAMAs) antimuscarinics, namely ipratropium and oxitropium, also block the inhibitory neuronal receptor M2, which potentially can cause vagally induced bronchoconstriction. Long acting antimuscarinic antagonists (LAMAs) such as tiotropium, aclidinium, glycopyrronium bromide and umeclidinium have prolonged binding to M3 muscarinic receptors, with faster dissociation from M2 muscarinic receptors, thus prolonging the duration of bronchodilator effect. Clinical trials have shown a greater effect on exacerbation rates for LAMA treatment versus LABA treatment [45].

Regular treatment with inhaled corticosteroids (ICS) alone does not modify the long-term decline of FEV_1 nor the mortality in patients with COPD. Glucocorticoids act at multiple points within the inflammatory cascade, although their effects in COPD are more modest as compared with bronchial asthma. Data from large patient studies suggest that inhaled corticosteroids can produce a small increase in post-bronchodilator FEV_1 and a small reduction in bronchial reactivity in stable COPD [46,47]. In patients with more advanced disease (usually classified as an FEV_1 <50% pred), there is evidence that the number of exacerbations per year and the rate of deterioration in health status can be reduced by inhaled corticosteroids in COPD. Evidence

from four large prospective 3-year studies has shown no effect of inhaled corticosteroids on rate of change of FEV_1 in any severity of COPD [48].

Theophylline exerts a small bronchodilator effect in stable COPD and it is associated with moderate symptomatic benefits [49].

Combination Bronchodilator Therapy

The GOLD ABCD tool combines symptom severity, using either the COPD Assessment Test score or the modified Medical Research Council scale, together with exacerbation risk, determined by either spirometry-defined airflow limitation or exacerbation history, to categorize patients into disease "risk stratification" groups ABCD to guide pharmacotherapy [50] (Figure 2).

The preference for long-acting muscarinic antagonist (LAMA)/long-acting β_2-agonist (LABA) combinations over inhaled corticosteroid (ICS)-containing regimens is supported by evidence from several studies. In the LANTERN and ILLUMINATE studies, a combination of glycopyrronium/indacaterol (LAMA/LABA) significantly improved lung function compared with salmeterol/fluticasone (LABA/ICS) and decreased the incidence of pneumonia in patients with moderate-to-severe COPD [51,52). Similarly, a LAMA/LABA combination of tiotropium/olodaterol provided a greater improvement in lung function than salmeterol/fluticasone in patients with moderate-to-severe COPD in the ENERGITO study [53]. In the FLAME study, glycopyrronium/indacaterol was more effective than salmeterol/fluticasone in reducing the rate of COPD exacerbations and increasing the time to first exacerbation in patients with a history of exacerbations in the previous year [54]. Notably, compared with LABA/ICS, LAMA/LABA combination therapy significantly reduced the rate of COPD exacerbations in patients with moderate-to-severe COPD who experienced either up to 1 or at least 1 exacerbation in the previous year.

If patients have persistent exacerbations despite being on the LAMA/LABA or LABA/ICS treatment regimens, LAMA/LABA/ICS triple therapy should be considered. A switch from LAMA/LABA to a triple therapy should be guided by the biomarker assessment (i.e., patients with eosinophil counts of ≥100 cells/μL are more likely to benefit from the triple therapy). The IMPACT Study (**I**nfor**M**ing the **Pa**thway of **C**OPD **T**reatment) has shown new evidence about the role of single inhaler triple therapy (ICS/LABA/LAMA) compared to ICS/LABA and LAMA/LABA. The main results of this study were obtained on reduction of exacerbation rate, lung function improvement (in terms of trough FEV_1 improvement), mortality data and incidence of pneumonia [55].

Other Anti-inflammatory Therapy for Stable COPD

Roflumilast is a selective inhibitor of the enzyme phosphodiesterase-4 (PDE-4), and targets systemic inflammation associated with COPD. Several clinical trials have reported benefit of roflumilast over placebo in patients with COPD in terms of FEV1 and exacerbations. Currently, there is no evidence for its use in patients with early disease, but only in severe and very severe COPD [56].

Macrolides have demonstrated a measurable ef-

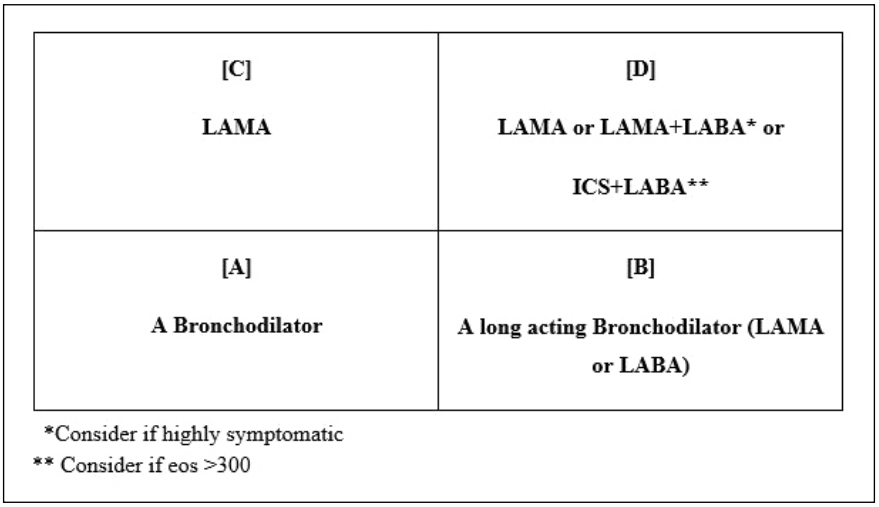

Figure 2. Pharmacological treatment recommendations based on 2022 GOLD Classification

ficacy in preventing exacerbations. However, their use in a chronic/preventive manner needs to be decided carefully balancing the potential efficacy in the right patients with the potential risk connected to an antibiotic overuse and potential antibiotic resistance in a single patient and/or a community [57].

Long-term use of oral corticosteroids has numerous side effects with no evidence of benefits. Regular treatment of mucolytics such as erdostein, carbocystein and NAC reduces the risk of exacerbations in selected populations. Furthermore, observational studies have shown that statins may have a positive effect on patients with COPD who receive them for cardiovascular and metabolic disease. Leukotriene modifiers have not been adequately tested in COPD patients. Finally, intravenous augmentation a1-antitrypsin therapy may slow down the progression of emphysema.

Mortality Data

Several studies have recently reported on the long-term mortality rate in COPD patients. The 5-year mortality of COPD patients was about 25.4%. Higher mortality was observed in males and the elderly. The 5-year mortality rate in males was about 1.5 times higher than in females. The common causes of death in COPD were chronic lower respiratory disease, lung cancer, cardiovascular disease, and cerebrovascular disease.

CONCLUSIONS

Early COPD remains poorly explored. The diagnosis of COPD as defined by currently established criteria indicates an established disease process which is irreversible. As no current pharmacological treatment is known to halt or reverse the progression of established COPD, it is essential for disease to be diagnosed early and prior to establishment of irreversible pathology, in order to allow timely interventions. Identification of pathological factors involved in the development of early disease will facilitate development of therapies targeting these early changes, and therefore potentially arrest or even reverse the disease process.

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REFERENCES

1. López-Campos JL, Tan W, Soriano JB. Global burden of COPD. Respirology. 2016;21(1):14-23.
2. Waatevik M, Skorge TD, Omenaas E, Bakke PS, Gulsvik A, Johannessen A. Increased prevalence of chronic obstructive pulmonary disease in a general population. Respir Med. 2013;107(7):1037-1045.
3. Agustí A, Hogg JC. Update on the Pathogenesis of Chronic Obstructive Pulmonary Disease. N Engl J Med. 2019;381(13):1248-1256.
4. Sampsonas F, Karkoulias K, Kaparianos A, Spiropoulos K. Genetics of chronic obstructive pulmonary disease, beyond a1-antitrypsin deficiency. Curr Med Chem. 2006;13(24):2857-2873.
5. Price DB, Tinkelman DG, Nordyke RJ, Isonaka S, Halbert RJ. Scoring system and clinical application of COPD diagnostic questionnaires. Chest. 2006;129(6):1531-1539.
6. Chan KPF, Ma TF, Kwok WC, Leung JKC, Chiang KY, Ho JCM, et al. Significant reduction in hospital admissions for acute exacerbation of chronic obstructive pulmonary disease in Hong Kong during coronavirus disease 2019 pandemic. Respir Med. 2020;171:106085.
7. Güder G, Brenner S, Angermann CE, Ertl G, Held M, Sachs AP, et al. "GOLD or lower limit of normal definition? A comparison with expert-based diagnosis of chronic obstructive pulmonary disease in a prospective cohort-study". Respir Res. 2012;13(1):13.
8. Grafino M, Todo-Bom F, Lutas AC, Cabral J, Pereira M, Valença J, et al. Spirometry for the diagnosis of airway obstruction in patients with risk factors for COPD: the GOLD and lower limit of normal criteria. J Bras Pneumol. 2022;47(6):e20210124.
9. Tzanakis N, Koulouris N, Dimakou K, Gourgoulianis K, Kosmas E, Chasapidou G, et al. Classification of COPD patients and compliance to recommended treatment in Greece according to GOLD 2017 report: the RELICO study. BMC Pulm Med. 2021;21(1):216.
10. Lange P, Halpin DM, O'Donnell DE, MacNee W. Diagnosis, assessment, and phenotyping of COPD: beyond FEV_1. Int J Chron Obstruct Pulmon Dis. 2016;11 Spec Iss(Spec Iss):3-12.
11. Peiffer G, Underner M, Perriot J. [COPD and smoking cessation: Patients' expectations and responses of health professionals]. Rev Pneumol Clin. 2018;74(6):375-390.
12. Anthonisen NR, Skeans MA, Wise RA, Manfreda J, Kanner RE, Connett JE. The effects of a smoking cessation intervention on 14.5-year mortality: a randomized clinical trial. Ann Intern Med. 2005;142(4):233-239.
13. Fletcher C, Peto R. The natural history of chronic airflow obstruction. Br Med J. 1977;1(6077):1645-1648.
14. Bafadhel M, Peterson S, De Blas MA, Calverley PM, Rennard SI, Richter K, et al. Predictors of exacerbation risk and response to budesonide in patients with chronic obstructive pulmonary disease: a post-hoc analysis of three randomised trials. Lancet Respir Med. 2018;6(2):117-126.
15. Di Marco F, Balbo P, de Blasio F, Cardaci V, Crimi N, Girbino G, et al. Early management of COPD: where are we now and

where do we go from here? A Delphi consensus project. Int J Chron Obstruct Pulmon Dis. 2019;14:353-360.
16. Tashkin DP. Smoking Cessation in Chronic Obstructive Pulmonary Disease. Semin Respir Crit Care Med. 2015;36(4):491-507.
17. Kalkhoran S, Glantz SA. E-cigarettes and smoking cessation in real-world and clinical settings: a systematic review and meta-analysis. Lancet Respir Med. 2016;4(2):116-128.
18. Froes F, Roche N, Blasi F. Pneumococcal vaccination and chronic respiratory diseases. Int J Chron Obstruct Pulmon Dis. 2017;12:3457-3468.
19. Kopsaftis Z, Wood-Baker R, Poole P. Influenza vaccine for chronic obstructive pulmonary disease (COPD). Cochrane Database Syst Rev. 2018;6(6):Cd002733.
20. Matsuse H. [Pneumococcal vaccine and influenza vaccine for prevention of exacerbation in patients with COPD]. Nihon Rinsho. 2016;74(5):839-842.
21. Blasi F, Bonanni P, Braido F, Gabutti G, Marchetti F, Centanni S. The unmet need for pertussis prevention in patients with chronic obstructive pulmonary disease in the Italian context. Hum Vaccin Immunother. 2020;16(2):340-348.
22. Cranston JM, Crockett AJ, Moss JR, Alpers JH. Domiciliary oxygen for chronic obstructive pulmonary disease. Cochrane Database Syst Rev. 2005;2005(4):Cd001744.
23. Ekström M, Ahmadi Z, Bornefalk-Hermansson A, Abernethy A, Currow D. Oxygen for breathlessness in patients with chronic obstructive pulmonary disease who do not qualify for home oxygen therapy. Cochrane Database Syst Rev. 2016;11(11):Cd006429.
24. Albert RK, Au DH, Blackford AL, Casaburi R, Cooper JA, Jr., Criner GJ, et al. A Randomized Trial of Long-Term Oxygen for COPD with Moderate Desaturation. N Engl J Med. 2016;375(17):1617-1627.
25. van der Leest S, Duiverman ML. High-intensity non-invasive ventilation in stable hypercapnic COPD: Evidence of efficacy and practical advice. Respirology. 2019;24(4):318-328.
26. Elliott MW, Nava S. Noninvasive ventilation for acute exacerbations of chronic obstructive pulmonary disease: "Don't think twice, it's alright!". Am J Respir Crit Care Med. 2012;185(2):121-123.
27. Chandra D, Stamm JA, Taylor B, Ramos RM, Satterwhite L, Krishnan JA, et al. Outcomes of noninvasive ventilation for acute exacerbations of chronic obstructive pulmonary disease in the United States, 1998-2008. Am J Respir Crit Care Med. 2012;185(2):152-159.
28. Marin JM, Soriano JB, Carrizo SJ, Boldova A, Celli BR. Outcomes in patients with chronic obstructive pulmonary disease and obstructive sleep apnea: the overlap syndrome. Am J Respir Crit Care Med. 2010;182(3):325-331.
29. Spruit MA, Singh SJ, Garvey C, ZuWallack R, Nici L, Rochester C, et al. An official American Thoracic Society/European Respiratory Society statement: key concepts and advances in pulmonary rehabilitation. Am J Respir Crit Care Med. 2013;188(8):e13-e64.
30. Donaldson GC, Wilkinson TM, Hurst JR, Perera WR, Wedzicha JA. Exacerbations and time spent outdoors in chronic obstructive pulmonary disease. Am J Respir Crit Care Med. 2005;171(5):446-452.
31. Jácome C, Marques A. Pulmonary rehabilitation for mild COPD: a systematic review. Respir Care. 2014;59(4):588-594.
32. Montes de Oca M, Loeb E, Torres SH, De Sanctis J, Hernández N, Tálamo C. Peripheral muscle alterations in non-COPD smokers. Chest. 2008;133(1):13-18.
33. Bourne S, DeVos R, North M, Chauhan A, Green B, Brown T, et al. Online versus face-to-face pulmonary rehabilitation for patients with chronic obstructive pulmonary disease: randomised controlled trial. BMJ Open. 2017;7(7):e014580.
34. Pompeo E, Mineo TC. Two-year improvement in multidimensional body mass index, airflow obstruction, dyspnea, and exercise capacity index after nonresectional lung volume reduction surgery in awake patients. Ann Thorac Surg. 2007;84(6):1862-1869; discussion -9.
35. Mineo TC, Pompeo E, Mineo D, Tacconi F, Marino M, Sabato AF. Awake nonresectional lung volume reduction surgery. Ann Surg. 2006;243(1):131-136.
36. Fessler HE, Permutt S. Lung volume reduction surgery and airflow limitation. Am J Respir Crit Care Med. 1998;157(3 Pt 1):715-722.
37. Sciurba FC, Rogers RM, Keenan RJ, Slivka WA, Gorcsan J, 3rd, Ferson PF, et al. Improvement in pulmonary function and elastic recoil after lung-reduction surgery for diffuse emphysema. N Engl J Med. 1996;334(17):1095-1099.
38. Ashraf O, Disilvio B, Young M, Ghosh S, Cheema T. Surgical Interventions for COPD. Crit Care Nurs Q. 2021;44(1):49-60.
39. Criner GJ, Cordova F, Sternberg AL, Martinez FJ. The National Emphysema Treatment Trial (NETT) Part II: Lessons learned about lung volume reduction surgery. Am J Respir Crit Care Med. 2011;184(8):881-893.
40. Stavem K, Bjørtuft Ø, Borgan Ø, Geiran O, Boe J. Lung transplantation in patients with chronic obstructive pulmonary disease in a national cohort is without obvious survival benefit. J Heart Lung Transplant. 2006;25(1):75-84.
41. Cazzola M, Page CP, Calzetta L, Matera MG. Pharmacology and therapeutics of bronchodilators. Pharmacol Rev. 2012;64(3):450-504.
42. Cazzola M, Matera MG. Bronchodilators: current and future. Clin Chest Med. 2014;35(1):191-201.
43. Higgins BG, Powell RM, Cooper S, Tattersfield AE. Effect of salbutamol and ipratropium bromide on airway calibre and bronchial reactivity in asthma and chronic bronchitis. Eur Respir J. 1991;4(4):415-420.
44. Melani AS. Long-acting muscarinic antagonists. Expert Rev Clin Pharmacol. 2015;8(4):479-501.
45. Bateman ED, Rennard S, Barnes PJ, Dicpinigaitis PV, Gosens R, Gross NJ, et al. Alternative mechanisms for tiotropium. Pulm Pharmacol Ther. 2009;22(6):533-542.
46. Burge PS, Calverley PM, Jones PW, Spencer S, Anderson JA, Maslen TK. Randomised, double blind, placebo controlled study of fluticasone propionate in patients with moderate to severe chronic obstructive pulmonary disease: the ISOLDE trial. Bmj. 2000;320(7245):1297-1303.
47. Pauwels RA, Löfdahl CG, Laitinen LA, Schouten JP, Postma

DS, Pride NB, et al. Long-term treatment with inhaled budesonide in persons with mild chronic obstructive pulmonary disease who continue smoking. European Respiratory Society Study on Chronic Obstructive Pulmonary Disease. N Engl J Med. 1999;340(25):1948-1953.
48. Vestbo J, Sørensen T, Lange P, Brix A, Torre P, Viskum K. Long-term effect of inhaled budesonide in mild and moderate chronic obstructive pulmonary disease: a randomised controlled trial. Lancet. 1999;353(9167):1819-1823.
49. Ram FS, Jones PW, Castro AA, De Brito JA, Atallah AN, Lacasse Y, et al. Oral theophylline for chronic obstructive pulmonary disease. Cochrane Database Syst Rev. 2002;2002(4):Cd003902.
50. Gupta N, Malhotra N, Ish P. GOLD 2021 guidelines for COPD - what's new and why. Adv Respir Med. 2021;89(3):344-346.
51. Zhong N, Wang C, Zhou X, Zhang N, Humphries M, Wang L, et al. LANTERN: a randomized study of QVA149 versus salmeterol/fluticasone combination in patients with COPD. Int J Chron Obstruct Pulmon Dis. 2015;10:1015-1026.
52. Vogelmeier CF, Bateman ED, Pallante J, Alagappan VK, D'Andrea P, Chen H, et al. Efficacy and safety of once-daily QVA149 compared with twice-daily salmeterol-fluticasone in patients with chronic obstructive pulmonary disease (ILLUMINATE): a randomised, double-blind, parallel group study. Lancet Respir Med. 2013;1(1):51-60.
53. Beeh KM, Derom E, Echave-Sustaeta J, Grönke L, Hamilton A, Zhai D, et al. The lung function profile of once-daily tiotropium and olodaterol via Respimat(®) is superior to that of twice-daily salmeterol and fluticasone propionate via Accuhaler(®) (ENERGITO(®) study). Int J Chron Obstruct Pulmon Dis. 2016;11:193-205.
54. Wedzicha JA, Banerji D, Chapman KR, Vestbo J, Roche N, Ayers RT, et al. Indacaterol-Glycopyrronium versus Salmeterol-Fluticasone for COPD. N Engl J Med. 2016;374(23):2222-2234.
55. Lipson DA, Barnhart F, Brealey N, Brooks J, Criner GJ, Day NC, et al. Once-Daily Single-Inhaler Triple versus Dual Therapy in Patients with COPD. N Engl J Med. 2018;378(18):1671-1680.
56. Wedzicha JA, Calverley PM, Rabe KF. Roflumilast: a review of its use in the treatment of COPD. Int J Chron Obstruct Pulmon Dis. 2016;11:81-90.
57. Taylor SP, Sellers E, Taylor BT. Azithromycin for the Prevention of COPD Exacerbations: The Good, Bad, and Ugly. Am J Med. 2015;128(12):1362.e1-e6.

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Immune checkpoint inhibitors and liver immune-related adverse effects: A comprehensive review of diagnosis and management

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Abstract

Immune checkpoint inhibitors (ICI) have greatly improved the management of advanced solid tumors with proven efficacy in overall survival rates. However, their increasing use is associated with a number of immune-mediated adverse events in almost every system organ. Liver toxicity, although rare, can be occasionally severe with the development of severe immune-mediated hepatitis. The current grading system assessing the severity of liver toxicity is suboptimal, overestimating the true incidence of severe hepatitis, and research should be guided towards this direction. Management includes the introduction of corticosteroids in cases of grade 2 or greater hepatitis but there are reports of spontaneous resolution without the use of immunosuppressive treatment. Thus, treatment algorithms need to be revised and predictive factors of spontaneous resolution need to be discovered. This review focuses on liver complications related to ICI treatment discussing incidence, diagnosis and treatment strategy currently used in this setting.

Key words: *Immune checkpoint inhibitors; Immune-mediated hepatitis; liver toxicity*

INTRODUCTION

In the last decade, the introduction of immune checkpoint inhibitors (ICIs) in the therapeutic management of different cancer types including unresectable or metastatic melanoma, advanced hepatocellular carcinoma and metastatic non-small cell lung cancer [1-4], has both changed treatment algorithms and improved overall survival rates in this setting. Since the approval of ipilimumab (anti-cytotoxic T-lymphocyte-associated protein 4 (CTLA4) antibody) for the treatment of unresectable or metastatic melanoma [3], many other ICIs have been introduced and their therapeutic indications have been expanded in other tumor types.

The molecular targets of ICIs are immune checkpoints, i.e., CTLA4, programmed cell death protein 1 (PD-1) and its ligands (PD-L1/PD-L2), which normally have an inhibitory effect on T-cell activation preventing auto-immunity, thus providing immunological tolerance to self-antigens [5, 6]. By inhibiting immune checkpoint signaling, T-cell activity is restored and the immune tolerance against specific tumor antigens is reversed thus promoting a durable anti-tumor immune response.

However, the abovementioned interference with the natural immunological tolerance can result in the development of various systemic toxicities due to the loss of T-cell inhibition leading to abnormal host immune responses [7]. These toxicities are called immune-related

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adverse events (irAEs) and can potentially affect every organ system (most commonly the skin, the gastrointestinal tract and the liver, and the endocrine system). They can mimic other immune-mediated disorders and have been associated with severe and sometimes fatal outcomes in cases of delayed diagnosis.

The aim of this review was to focus on the immune-related hepatic complications resulting from ICI treatment as well as its management, which is mostly based on expert opinions. Due to the lack of randomized controlled trials on this topic, it was impossible to conduct a systematic review.

METHODS

MEDLINE databases were searched for eligible studies from August 2006 to April 2022 using the textwords "Checkpoint inhibitors" or "immune checkpoint inhibitors" or "anti-CTLA4" or "anti-CTLA-4" or "anti-PD1" or "anti-PDL1" or "ipilimumab", or "pembrolizumab" or "nivolumab" and "hepatitis" or "liver toxicity" or "toxicity" or "liver" or" adverse event" or "immune-related adverse event". All relevant review articles (English) were manually searched, and all original studies were retrieved from them.

Epidemiology of liver irAEs

The overall incidence of irAEs is ranging between 15% and 90%, and as already mentioned, every organ system can be potentially affected [7-9]. The dermatological and gastrointestinal (colon, small intestine, liver and pancreas) irAEs are the most common toxicities [10-12]. The incidence and severity of irAEs seem to be unaffected by the tumor type [13]. However, CTLA inhibitors are more frequently associated with irAEs compared to anti-PD1/PDL1 agents, and these toxicities are usually more severe. In a meta-analysis [8] of 1265 patients from 22 clinical trials, the overall incidence of all-grade irAEs in oncologic patients receiving anti-CTLA4 antibodies (ipilimumab and tremelimumab) was 72 % (95 % CI, 65–79 %) (high-grade irAEs, 24 % (95 % CI, 18–30 %)); this association was found to be dose-dependent. In a systematic review of 23 studies comprising 3284 patients in the PD-1 group and 2460 patients in the PD-L1 group [14], the overall incidence of all-grade adverse events was 64% (95% CI, 63%-66%]) and 66% (95% CI, 65%-69%) for PD-1 and PD-L1 inhibitors ((high-grade irAEs, 13% (95% CI, 12%-14%) and 21% (95% CI, 19%-23%)), respectively. The risk increases substantially for patients treated with both CTLA4 and PD1/PDL1 inhibitors, implying an additive toxicity when combination is used [15]. On the other hand, in a retrospective study [16] which aimed to assess the safety profile of nivolumab monotherapy in patients with advanced melanoma, the objective response rate was significantly higher in patients that demonstrated irAEs compared to those that did not (48.6% s 17.8%, $p<0.001$).

Regarding liver irAEs, the incidence of all-grade and high-grade immune-mediated hepatitis in ICI users is around 5% and 1-2%, respectively [17]. This incidence increases at 25% and 8-10%, respectively, when combination of ICIs is used [15, 18]. It seems that the risk is higher in ICI-users treated for hepatocellular carcinoma compared to those treated for non-liver cancers [19].

Dermatological irAEs usually occur at around four weeks after initiation of treatment, together with gastrointestinal ones (at around six weeks), whereas liver toxicity appears later at approximately 8-12 weeks after starting ICI treatment [16]. However, liver abnormalities, as all other irAEs, can occur even after longer time periods following initial administration [20].

Grading of severity of liver irAEs

The severity of irAEs is more commonly graded using the US National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE) [21]. Depending on these criteria, the severity is graded on a scale of 1 to 5 as follows: grade 1 (mild toxicity), grade 2 (moderate toxicity), grade 3 (severe toxicity), grade 4 (life-threatening) and grade 5 (death). The grading of severity of ICI-related hepatitis is depicted in Table 1.

This grading scale has the advantage of stratifying treatment management and thus, it has been extensively used in clinical trials of ICIs allowing comparisons between studies; however, it is accompanied by several drawbacks, most importantly the overestimation of the incidence and severity of symptoms by physicians [6]. According to the European Association for the Study of the Liver (EASL) guidelines [22], drug-induced liver injury is considered severe if the elevation of transaminases is accompanied with an increase in bilirubin (Hy's law), whereas the level of elevation of liver enzymes alone is not sufficient to reflect the severity of liver injury. This is not in accordance with CTCAE criteria [21], where grade 4 hepatotoxicity is defined as very high levels of transaminases without concomitant increase in bilirubin. Consequently, the CTCAE grading system is considered suboptimal and more research is needed in the setting of severity stratification.

Table 1. *Grading of severity of ICI hepatitis (National Cancer Institute's Common Terminology Criteria for Adverse Events (CTCAE), version 5.0).*

Adverse Event	Grade 1	Grade 2	Grade 3	Grade 4	Grade 5
Hepatitis	AST or ALT 1 3×ULN and/or T-BIL 1 1.5×ULN	AST or ALT 3 5×ULN and/or T-BIL 1.5 3×ULN	AST or ALT 5-20×ULN and/or T-BIL 3-10×ULN	AST or ALT >20×ULN and/or T-BIL >10×ULN	Death

AST: aspartate aminotransferase, ALT: alanine aminotransferase, t-bil: total bilirubin, ULN: Upper Limit of normal

Clinical presentation of liver irAEs

ICI hepatitis is mostly asymptomatic and usually follows a hepatocellular pattern of liver injury characterized by elevations of aminotransferases (ALT and AST) with or without mildly increased total bilirubin levels [23-25]. Cases of cholestatic liver injury have been reported but they are not the rule [26]. Symptoms like fever, malaise, abdominal discomfort or jaundice are rare. Moreover, fulminant hepatitis causing acute liver failure is very uncommon with an incidence of about 0.4% [27]. Taking the abovementioned into account, the diagnosis of ICI-hepatitis is mostly incidental following routine blood testing.

Although an immune-mediated hepatitis, ICI hepatitis should be dissociated from autoimmune hepatitis. The histological features of ICI hepatitis are usually portal and periportal hepatitis and hepatocellular necrosis (mostly centrilobular) with infiltration by lymphocytes, plasma cells and eosinophils, thus resembling acute viral or autoimmune hepatitis [28, 29]. However, high titles of anti-nuclear antibodies (ANA), anti-smooth muscle antibodies (ASMA) and other autoimmune hepatitis-related auto-antibodies are usually absent and ICI hepatitis typically responds to drug discontinuation and one course of immunosuppressive treatment without recurrence, thus is differentiated from idiopathic autoimmune hepatitis [30, 31]. Furthermore, the presence of cirrhosis, emperipolesis and rosette formation are hallmarks of autoimmune hepatitis, not typical in ICI-related hepatitis [22, 31]. Interestingly, one histological pattern specific for patients under treatment with anti-CTLA4 agents is that of non-necrotizing granulomatous hepatitis, whereas liver injury by anti-PD1/PDL1 has a more heterogenous pattern without the presence of granulomas [23, 32]. Bile duct injury with the presence of cholangitis in histological specimens following treatment with ICIs has also been reported and is usually mild [33]. Other unusual presentations demonstrated in case reports are sclerosing cholangitis, nodular regenerative hyperplasia, sinusoidal obstruction syndrome and vanishing bile duct syndrome [34].

Diagnosis of liver irAEs

The diagnosis of ICI-related hepatitis is mostly a diagnosis of exclusion. The initial approach includes detailed medical history (including alcohol and concomitant drugs/herbs use) and physical examination. Infectious causes of abnormal liver function tests (i.e., viral hepatitis A, B, C, and E, Epstein-Barr virus, Cytomegalovirus, Herpes Simplex virus) should be excluded [22, 35-37]. Auto-antibodies including ANA, ASMA, anti-mitochondrial antibodies (if cholestatic injury pattern), liver-kidney microsomal type 1 (LKM-1) antibodies, as well as quantitative assessment of immunoglobulins should be assessed. An abdominal ultrasound is also part of the initial work-up to exclude vascular thrombosis, hepatic metastases, liver cirrhosis or biliary obstruction.

The question is if liver biopsy is indicated in the initial assessment of patients under ICI treatment presenting with abnormal liver function tests [19]. As mentioned before, liver histology can assist in both the diagnosis of ICI-related liver toxicity and the assessment of severity of liver injury; thus, it can guide treatment management. Currently, liver biopsy is reserved for patients with grade 3 or greater liver toxicity and/or to exclude alternative diagnoses.

Management of liver irAEs

According to the EASL recommendations [22], which rely on clinical experience and the management of autoimmune hepatitis, before the initiation of treatment with ICIs, baseline liver parameters and the patient's lipid profile should be assessed. Potential confounding factors such as pre-existing liver diseases and presence of liver metastases, as well as viral infections (HIV, HBV, HCV, HEV) should be excluded. Underlying autoimmune hepatitis or other autoimmune disorders should be investigated.

Following initiation of treatment, liver function tests

should be monitored every two weeks for the first 8 to 12 weeks of treatment, and then every four weeks [22].

According to the Multinational Association of Supportive Care in Cancer (MASCC) 2020 clinical practice recommendations [36] and the American Society of Clinical Oncology (ASCO) Clinical Practice Guidelines [35], in cases of grade 1 liver toxicity, ICI treatment can be continued but with careful blood monitoring (AST, ALT, total bilirubin) 1-2 times/week. No specific treatment is recommended in this stage apart from supportive care for symptomatic control.

ICI treatment should be temporarily discontinued if grade 2 or 3 liver toxicity, but permanently withdrawn if grade 4. In the case of grade 2 or greater hepatitis, corticosteroids should be initiated if the abnormal liver tests persist or worsen with significant clinical symptoms in 3 to 5 days. The recommended dose is 0.5-1.0 mg/kg/day prednisone or equivalent, 1-2 mg/kg/d methylprednisolone or equivalent, and 2 mg/kg/d methylprednisolone equivalents if grade 2, 3 and 4 hepatitis, respectively. Steroids should be tapered over 6-8 weeks. ICI treatment can be resumed (if grade 2/3 hepatitis) when liver parameters improve to grade 1 or normal values while on prednisone ≤ 10 mg/day (or equivalent).

In patients with hepatitis refractory to corticosteroids or no responsiveness after three days of continuous administration, a second immunosuppressive regimen should be added. According to ASCO guidelines [35], mycophenolate mofetil (MMF) (500 – 1000 mg BID) or azathioprine (1–2 mg/kg) or tacrolimus (targeting blood levels of 8–10 ng/ml or lower in case of an early response) is recommended. The role of infliximab in immune-mediated hepatitis is unclear considering the potential hepatotoxicity associated with infliximab use. Antithymocyte globulin (1.5 mg/kg) for 48 hours has been added to the treatment with MMF and steroids [38] in cases of severe, fulminant hepatitis and has been reported to be effective.

According to recent data [23], not all patients experiencing immune-mediated hepatitis following treatment with ICI need corticosteroids. More specifically, 16 out of 536 patients treated with anti-PD-1/PD-L1 or CTLA-4 immunotherapy developed histologically proven immune-mediated hepatitis. The decision to start steroids was based on biological (bilirubin >2.5 mg/dl and/or international normalized ratio [INR] >1.5) and/or histological criteria for severity assessment. Overall, six patients presented spontaneous resolution of hepatitis without receiving any corticosteroid treatment. Furthermore, in three of these patients, ICI treatment was re-introduced without recurrence of liver toxicity. The abovementioned indicate again that further investigation is needed regarding severity stratification and accordingly, treatment management in patients presenting with liver irAEs.

CONCLUSIONS

Despite the breakthrough in the management of advanced solid tumors after the introduction of checkpoint inhibitors, a variety of immune-mediated toxicities have emerged. These can be occasionally severe; thus, physicians should be aware of these entities in order to identify them early and to treat them appropriately. Liver irAEs are rare; however, their incidence increases substantially following combination treatment with ICIs. There is a knowledge gap regarding the pathophysiology of these toxicities and the specific risk factors for these adverse events, if any, are not yet elucidated. Better algorithms to identify patients in need for initiation of steroids are needed as well as prognostic indicators for treatment response and recurrence. The diagnosis of liver irAEs is problematic and research should be guided towards the identification of specific biomarkers and/or diagnostic tools to assist with the differential diagnosis of immune-mediated hepatitis. Lastly, the grade classification system should be reviewed and revised to better stratify the grade of severity from a hepatologist's perspective.

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REFERENCES

1. Pardoll DM. The blockade of immune checkpoints in cancer immunotherapy. Nat Rev Cancer 2012;12(4):252–64.
2. Finn RS, Qin S, Ikeda M, Galle PR, Ducreux M, Kim TY, et al. Atezolizumab plus bevacizumab in unresectable hepatocellular carcinoma. N Engl J Med 2020;382(20):1894–905.
3. Hodi FS, O'Day SJ, McDermott DF, Weber WR, Sosman JA, Haanenet JB, al. Improved survival with ipilimumab in patients with metastatic melanoma. N. Engl J Med 2010;363(8):711–23.
4. Adachi K, Tamada, K. Immune checkpoint blockade opens

an avenue of cancer immunotherapy with a potent clinical efficacy. Cancer Sci 2015;106(8):945–50.
5. Okoye IS, Houghton M, Tyrrell L, Barakat K, Elahi S. Coinhibitory receptor expression and immune checkpoint blockade: maintaining a balance in CD8(+) T cell responses to chronic viral infections and cancer. Front Immunol 2017;8:1215.
6. Samaan MA, Pavlidis P, Papa S, Powell N, Irving PM. Gastrointestinal toxicity of immune checkpoint inhibitors: from mechanisms to management. Nat Rev Gastroenterol Hepatol 2018;15(4):222-34.
7. Villadolid J, Amin A. Immune checkpoint inhibitors in clinical practice: update on management of immune-related toxicities. Transl Lung Cancer Res 2015;4(5):560–75.
8. Bertrand A, Kostine M, Barnetche T, Truchetet ME, Schaeverbeke T. Immune related adverse events associated with anti-CTLA-4 antibodies: systematic review and meta-analysis. BMC Med 2015;13:211.
9. Maughan BL, Bailey E, Gill DM, Agarwal N. Incidence of Immune-Related Adverse Events with Program Death Receptor-1- and Program Death Receptor-1 Ligand-Directed Therapies in Genitourinary Cancers. Front Oncol 2017;7:56.
10. Xu C, Chen YP, Du XJ, Liu JQ, Huang CL, Chen L, et al. Comparative safety of immune checkpoint inhibitors in cancer: systematic review and network metaanalysis. BMJ 2018;363:k4226.
11. Weber JS, Dummer R, de Pril V, Lebbe C, Hodi FS, MDX010-20 Investigators. Patterns of onset and resolution of immune-related adverse events of special interest with ipilimumab: detailed safety analysis from a phase 3 trial in patients with advanced melanoma. Cancer 2013;119(9):1675-682.
12. Costa R, Carneiro BA, Agulnik M, Rademaker AW, Pai SG, Villaflor VM, et al. Toxicity profile of approved anti-PD-1 monoclonal antibodies in solid tumors: a systematic review and meta-analysis of randomized clinical trials. Oncotarget 2017;8(5):8910-20.
13. Bourke JM, O'Sullivan M, Khattak MA. Management of adverse events related to new cancer immunotherapy (immune checkpoint inhibitors). Med J Aust 2016;205(9):418-24.
14. Pillai RN, Behera M, Owonikoko TK, Kamphorst AO, Pakkala S, Belan CP, et al. Comparison of the toxicity profile of PD-1 versus PD-L1 inhibitors in non-small cell lung cancer: A systematic analysis of the literature. Cancer 2018;124(2):271-7.
15. Xing P, Zhang F, Wang G, Xu Y, Li C, Wang S, et al. Incidence rates of immune-related adverse events and their correlation with response in advanced solid tumours treated with NIVO or NIVO+IPI: a systematic review and meta-analysis. J Immunother Cancer 2019;7(1):341.
16. Weber JS, Hodi FS, Wolchok JD, Topalian SL, Schadendorf D, Larkin J, et al. Safety Profile of Nivolumab Monotherapy: A Pooled Analysis of Patients with Advanced Melanoma. J Clin Oncol 2017;35(7):785-92.
17. Dougan M. Gastrointestinal and Hepatic Complications of Immunotherapy: Current Management and Future Perspectives. Curr Gastroenterol Rep 2020;22(4):15.
18. Shivaji UN, Jeffery L, Gui X, Smith SCL, Ahmad OF, Akbar A, et al. Immune checkpoint inhibitor-associated gastrointestinal and hepatic adverse events and their management. Therap Adv Gastroenterol 2019;12:1756284819884196.
19. De Martin E, Michot JM, Rosmorduc O, Guettier C, Samuel D. Liver toxicity as a limiting factor to the increasing use of immune checkpoint inhibitors. JHEP Rep 2020;2(6):100170.
20. Parakh S, Cebon J, Klein O. Delayed autoimmune toxicity occurring several months after cessation of anti-PD-1 therapy. Oncologist 2018;23(7):849–51.
21. US Department of Health and Human Services. Common Terminology Criteria for Adverse Events (CTCAE) version 4.0. http://evs.nci.nih.gov/ftp1/CTCAE/CTCAE_4.03_2010-06-14_QuickReference_8.5x11.pdf
22. European Association for the Study of the Liver. EASL Clinical Practice Guidelines: Drug-induced liver injury. J Hepatol 2019;70(6):1222-261.
23. De Martin E, Michot J-M, Papouin B, Champiat S, Mateus C, Lambotte O, et al. Characterization of liver injury induced by cancer immunotherapy using immune checkpoint inhibitors. J Hepatol 2018;68(6):1181–90.
24. Riveiro-Barciela M, Barreira-Díaz A, Vidal-González J, Muñoz-Couselo E, Martínez-Valle F, Viladomiu L, et al. Immune-related hepatitis related to checkpoint inhibitors: clinical and prognostic factors. Liver Int 2020; 40(8):1906-16.
25. Huffman BM, Kottschade LA, Kamath PS, Markovic SN. Hepatotoxicity after immune checkpoint inhibitor therapy in melanoma: natural progression and management. Am J Clin Oncol 2018;41(8):760–5.
26. Cheung V, Gupta T, Payne M, Middleton MR, Collier JD, Simmons A, et al. Immunotherapy-related hepatitis: real-world experience from a tertiary centre. Frontline Gastroenterol 2019;10(4):364–71.
27. Wang DY, Salem J-E, Cohen JV, Chandra S, Menzer C, Ye F, et al. Fatal toxic effects associated with immune checkpoint inhibitors: a systematic review and meta-analysis. JAMA Oncol 2018;4(12):1721–8.
28. Johncilla M, Misdraji J, Pratt DS, Agoston AT, Lauwers GY, Srivastava A, et al. Ipilimumab-associated hepatitis: clinicopathologic characterization in a series of 11 cases. Am J Surg Pathol 2015;39(8):1075–84.
29. Papouin B, Mussini C, De Martin E, Guettier C. Hepatic and digestive adverse events of immune checkpoint inhibitors (anti-CTLA-4 and anti-PD-1/PD-L1): a clinico-pathological review. Ann Pathol 2018;38(6):338–51.
30. Champiat S, Lambotte O, Barreau E, Belkhir R, Berdelou A, Carbonnelet F, et al. Management of immune checkpoint blockade dysimmune toxicities: a collaborative position paper. Ann Oncol 2016;27(4):559–74.
31. Belli C, Zuin M, Mazzarella L, Trapani D, D'Amico P, Guerini-Rocco E, et al. Liver toxicity in the era of immune checkpoint inhibitors: A practical approach. Crit Rev Oncol Hematol 2018;132:125-9.
32. Kleiner DE, Berman D. Pathologic changes in ipilimumab-related hepatitis in patients with metastatic melanoma. Dig Dis Sci 2012;57(8):2233–40.
33. Zen Y, Yeh MM. Hepatotoxicity of immune checkpoint inhibitors: a histology study of seven cases in comparison

with autoimmune hepatitis and idiosyncratic drug-induced liver injury. Mod Pathol 2018;31(6):965–73.
34. Fessas P, Possamai LA, Clark J, Daniels E, Gudd C, Mullish BH, et al. Immunotoxicity from checkpoint inhibitor therapy: clinical features and underlying mechanisms. Immunology 2020;159(2):167-77.
35. Brahmer JR, Lacchetti C, Schneider BJ, Atkins MB, Brassil KJ, Caterino JM, et al. Management of Immune-Related Adverse Events in Patients Treated With Immune Checkpoint Inhibitor Therapy: American Society of Clinical Oncology Clinical Practice Guideline. J Clin Oncol2018;36(17):1714-68.
36. Dougan M, Blidner AG, Choi J, Cooksley T, Glezerman I, Ginex P, et al. Multinational Association of Supportive Care in Cancer (MASCC) 2020 clinical practice recommendations for the management of severe gastrointestinal and hepatic toxicities from checkpoint inhibitors. Support Care Cancer 2020;28(12):6129-43.
37. Haanen J, Carbonnel F, Robert C, Kerr KM, Peters S, Larkin J, et al. Management of toxicities from immunotherapy: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. Ann Oncol 2017;28:iv119–42.
38. Boutros C, Tarhini A, Routier E, Lambotte O, Ladurie FL, Carbonnel F, et al. Safety profiles of anti-CTLA-4 and anti-PD-1 antibodies alone and in combination. Nat Rev Clin Oncol 2016;13(8):473-86.

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Recognizing medical emergencies- Hyperthermia

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Abstract

Hyperthermia is defined by the elevation of core body temperature above 40,5 °C, due to primary dysfunction of the thermoregulatory center in hypothalamus. It is considered a medical emergency that can lead to multi-organ dysfunction and death if not treated promptly and appropriately. The differential diagnosis is wide and the emergency medicine physicians should be aware of the main causes of febrile illness which include, except from infections and sepsis, intoxication from medication and illicit substances, neuroleptic malignant syndrome, malignant hyperthermia, endocrine disorders and environmental-related conditions. The primary goal of treatment is the decrease of core body temperature. The main cooling techniques are simple to use and should be applied in the Emergency Department immediately after the diagnosis of the condition, along with disease-specific treatment. Early recognition of the cause of hyperthermia can lead to improved outcomes in morbidity and mortality. In this topic, some of the main causes of hyperthermia will be discussed.

Key words: *Hyperthermia; drug intoxication; heat stroke; cooling techniques; surface cooling*

INTRODUCTION

Normal body temperature is approximately 37°C (degrees Celsius) or 98.6°F (degrees Fahrenheit) and varies by about 0.5°C during the day. Fever is core body temperature elevation above a "set-point" which is controlled by the thermoregulatory center in hypothalamus. This, usually, happens as a response to cytokines activated by infection or sterile inflammation [1]. On the other side, hyperthermia is a medical emergency due to complete loss of thermal control by the thermoregulatory system, leading to excessive heat generation and multi-system dysfunction [2]. The differentiating features between fever and hyperthermia can be seen in Table 1.

Core temperature (Tc) reflects the temperature of the internal organs and best describes an individual's thermal status. The measurement of Tc can be made in many sites (Table 2), but four sites are considered to give more accurate measurements of Tc, the tympanic membrane, nasopharynx, esophagus and pulmonary artery. In clinical settings the esophagus measurements are considered to be the gold standard whereas the tympanic measurements provide an alternative non-invasive technique [3].

The most common clinical manifestations of hyperthermic patients are tachypnea, tachycardia and hypotension. The skin is usually warm and in some cases sweating can be absent (hot and dry skin) [2]. Nearly every system can be impaired and the patients can present with neurologic dysfunction, such as delirium, seizures, coma etc., cardiogenic or noncardiogenic pulmonary edema, other cardiac manifestations like arrhythmias or ischemic changes, acute respiratory distress syndrome (ARDS), acute kidney injury, electrolyte and acid-base disturbances, hepatic dysfunction

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Table 1. *Differentiating features between fever and hyperthermia.*

	Fever	Hyperthermia
Temperature	Usually < 41C	>41 °C suggests hyperthermia >41,5 °C strongly indicative
Effect of antipyretics	Effective	No effect
Motor features	Rigors	Rigidity or clonus more frequently related to some forms of medication-induced hyperthermia

Table 2. *Summary of temperature measurements.*

Measurement location	Accuracy	Advantages	Disadvantages
Body surface	Low	Easy and widely available	Inaccurate
Oral	Low	Easy and widely available	Inaccurate
Axilla	Low	Easy and widely available	Inaccurate
Tympanic membrane	Satisfactory	Precise, repeatable and brain core temperature	High risk of measurement error
Rectum	Satisfactory	Easy, widely available, precise and repeatable	High latency
Urinary bladder	Satisfactory	precise and repeatable	High latency
Nasopharynx	Satisfactory	Easy, widely available	High risk of measurement error
Esophagus	Most accurate	Easy, widely available and repeatable	High latency
Pulmonary artery	Most accurate	precise and repeatable	Invasive and restricted to intensive care units

and gastrointestinal hemorrhage, rhabdomyolysis and coagulopathy or even disseminated intravascular coagulation (DIC) [4].

Causes of hyperthermia

Elevated Tc is one of the most frequently recorded vital signs among Emergency Department (ED) patients. It is difficult to differentiate between fever and hyperthermia. Fever is the main cause of elevated body temperature and is usually attributed to infection, complicated or not by sepsis or septic shock. However, if the patient's condition does not improve after the administration of fluids and antibiotics, sepsis mimics should be considered as the cause of elevated Tc. Some of these clinical conditions can be life-threatening if not treated early and appropriately. It is of great importance to identify the causes of hyperthermia (Table 3) in order to improve ED patients' outcomes [5].

1. Neuroleptic malignant syndrome

Neuroleptic malignant syndrome (NMS) is a life-threatening syndrome and it is characterized by altered mental status, hyperthermia, autonomic instability and muscle rigidity ("lead pipe" rigidity). It is associated with the use of dopamine-receptor antagonist medications but can also be precipitated by withdrawal from dopaminergic medications (e.g. those used in the treatment of Parkinson's disease). It is usually reported with the typical antipsychotics, like haloperidol and fluphenazine, but it can be caused by nearly every neuroleptic medication (including antiemetics like droperidol, metoclopramide etc). Risk factors for the development of NMS are high doses of medication, rapid escalation, and parenteral use. The syndrome can occur anytime during the course of treatment, but it usually develops within the first two weeks of the initiation of the neuroleptic agent [5][6]. The diagnosis is based on DSM-V (diagnostic and statistical manual for mental disorders) criteria:

Table 3. *Differential diagnosis of hyperthermia.*

Infection	Drug or toxin related	Neurologic	Environmental	Endocrine	Oncologic
Sepsis	Malignant hyperthermia	Hypothalamic stroke	Heat-related illnesses	Thyroid storm	Lymphoma
Meningitis	Neuroleptic malignant syndrome	Status epilepticus		Pheochromocytoma/ paraganglioma	Leukemia
Encephalitis	Withdrawal syndromes (e.g. alcohol, benzodiazepines etc.)	Intracerebral hemorrhage		Diabetic ketoacidosis	
Brain abscess	Illicit drugs (cocaine)				
Tetanus	Sympathomimetic intoxication (e.g.amphetamines)				
Typhoid fever	Anticholinergic intoxication (e.g. antihistamine)				
Malaria	Serotonin syndrome				
	Salicylate poisoning				

Major criteria (all required)

- Exposure to dopamine-blocking agent
- Severe muscle rigidity
- Fever

Other Criteria (at least two required)

- Diaphoresis
- Dysphagia
- Tremor
- Incontinence
- Altered level of consciousness
- Mutism
- Tachycardia
- Elevated or labile blood pressure
- Leukocytosis

Elevated creatine phosphokinase [6]

The standard treatment pathway for NMS comprises the discontinuation of the causative agent and supportive care including rehydration, support of the cardiopulmonary system, maintenance of normothermia and prevention of complications (e.g. heparin for deep vein thrombosis prophylaxis). Benzodiazepines, lorazepam or diazepam, can be used to control agitation if necessary. Empiric pharmacological treatment, such as bromocriptine or amantadine orally or dantrolene intravenously, has been used in more severe cases. Electroconvulsive therapy has occasionally been used in some refractory cases. ED physicians' awareness and early detection of the syndrome are crucial for the prognosis. Delayed treatment can lead to increased morbidity and fatality [6,7].

2. Serotonin syndrome

Serotonin syndrome (SS) is a potentially life-threatening condition which is frequently misdiagnosed. The most commonly implicated medications are SSRIs (Selective Serotonin Reuptake Inhibitors), linezolid and fentanyl. It can be precipitated by therapeutic medication use or intentional self-poisoning and is more frequently seen with the co-administration of the above agents [5]. The clinical presentation of SS includes autonomic instability, such as tachycardia, hyperthermia and blood pressure disturbances, neuromuscular excitation (i.e. hyperreflexia, tremor, clonus) and mental status changes like agitation, delirium and coma [8]. The physical examination can reveal other signs, such as dilated pupils, flushed skin and diaphoresis, dry mucous membranes and bilateral Babinski sign [9]. The diagnosis is based on the Hunter criteria:

Use of a serotonergic agent plus one of the following:

- Spontaneous clonus
- Inducible clonus and agitation or diaphoresis
- Ocular clonus and agitation or diaphoresis
- Tremor and hyperreflexia
- Hypertonia

Temperature >38°C and ocular clonus or inducible clonus [10]

The primary components of the emergent treatment of SS are the discontinuation of the offending medication and supportive care. The administration of intravenous (IV) fluids and oxygen, as well as the continuous cardiac monitoring and the correction of

vital signs' abnormalities constitute the mainstay of therapy. Sedation with benzodiazepines (IV lorazepam or diazepam) is used for agitation control. If agitation does not improve with the above measures cyproheptadine, a histamine-1 receptor antagonist, is suggested as an antidote [11]. If Tc is above 41,1°C, immediate sedation, paralysis, and endotracheal intubation are necessary along with the standard cooling techniques. Prognosis is in general favorable and unlike NMS (Table 4), most cases with mild to moderate symptoms resolve within 24 hours [9].

3. Malignant Hyperthermia

Malignant hyperthermia (MH) is a progressive, hypermetabolic, life-threatening reaction caused by the exposure to a volatile anesthetic (e.g. halothane, isoflurane etc.) or succinylcholine. A genetic disorder of skeletal muscle receptors is the abnormality that leads MH-susceptible persons receiving general anesthesia to result in excessive muscle contraction, rhabdomyolysis, anaerobic metabolism and acidosis [12,13]. The most frequent initial clinical signs that usually develop within minutes after the administration of the offending anesthetic, are hypercarbia (elevated end-tidal CO2), tachypnea and sinus tachycardia. Hyperthermia often occurs later on the course of MH reaction, along with generalized muscle rigidity, masseter muscle rigidity and arrhythmias [14,15]. Treatment of MH consists of the immediate discontinuation of the anesthetic, the optimization of oxygenation and ventilation, management of cardiac arrhythmias, the administration of intravenous dantrolene and correction of laboratory and acid-base abnormalities. Cooling techniques can be used for patients with Tc above 39 °C [16]. Awareness should be raised among emergency medicine physicians to the possibility of fatal non-anesthesia triggered MH episodes in MH-susceptible persons. People with the genetic disorder can present with similar to MH clinical features after exposure to thermal stress, including febrile illness or exercise. This disorder is characterized as "non-anesthetic" or "awake" malignant hyperthermia and it is usually resolved spontaneously or after the administration of low dose of oral dantrolene (i.e. 25 mg orally) [17,18].

4. Salicylate toxicity

The best-known salicylate is acetylsalicylic acid (aspirin). Patients with salicylate toxicity will present with high body temperature, tachycardia, tachypnea and lactic acidosis. Acidosis is caused by the interference of salicylate with the anaerobic metabolism. The acid-base changes that are observed in these cases are firstly respiratory alkalosis due to centrally mediated hyperventilation, subsequently compensatory non-gap metabolic acidosis and finally anion-gap metabolic acidosis due to lactate accumulation. The classic triad of symptoms includes hyperpnea, tinnitus and gastrointestinal irritation. With the progression of metabolic acidosis, the patient deteriorates, and severe complications arise such as pulmonary edema, CNS (Central Nervous System) depression and even death. Aggressive treatment may be needed in the emergency department like volume resuscitation, airway management, systemic alkalinization with sodium bicarbonate and hemodialysis [5,19,20].

5. Anticholinergic toxicity

Anticholinergics are among the most frequently reported drugs that can be associated with non-pyrogenic hyperthermia and heat-related mortality in the elderly [21]. They can interfere with the ability to sweat by the inhibition of muscarinic receptors on the sweat glands resulting in anhidrotic hyperthermia [5]. Antihistamines, tricyclic antidepressants and recreational drugs can lead to the development of an anticholinergic toxidrome with dry mucous membranes, dilated unreactive pupils, paralytic ileus and urinary retention, as long as symptoms from the CNS like agitation that can progress to

Table 4. *Distinguishing features between NMS and SS.*

	Serotonin syndrome (SS)	Neuroleptic Malignant syndrome (NMS)
Onset	Within 24 h	Days to weeks
Neuromuscular findings	Hyperreactivity (tremor, clonus)	Bradyreflexia, severe muscular rigidity
Causative agents	Serotonin agonists	Dopamine antagonists
Treatment agents	Benzodiazepines, cyproheptadine	Bromocriptine, dantrolene
Resolution	Within 24 h	Days to weeks

delirium [22]. Physostigmine, a cholinesterase inhibitor, is used as an antidote for the treatment of anticholinergic delirium. Benzodiazepines can be used for agitation and seizures along with supportive care [5,22].

6. Sympathomimetic toxicity or withdrawal from sympathetic antagonists

Illicit sympathomimetic drugs, such as cocaine, amphetamines, methylenedioxymethamphetamine (MDMA), can cause severe toxicity due to increased adrenergic response. Hypertension, tachycardia, dilated but reactive pupils (unlike anticholinergic toxicity) (Table 5), psychomotor agitation and hyperthermia. The degree of hyperthermia is related to the mortality rate. The goal of treatment is to control the excessive adrenergic stimulation and agitation. Benzodiazepines are the mainstay of treatment, and they are titrated to the desired effect. In more severe cases, the addition of a nondepolarizing neuromuscular blocker may be necessary. Cooling techniques for passive and active cooling, depending on the severity of hyperthermia, can be used for further thermal control [23].

Withdrawal from alcohol or benzodiazepines can result in excessive sympathetic activity. Symptoms from benzodiazepines withdrawal usually develop between 2-10 days after the discontinuation of the drug, while symptoms from alcohol withdrawal manifest within the first 48-72 hours since the last alcohol consumption [24]. Clinical manifestations of benzodiazepine withdrawal vary from mild symptoms, like muscle-spasms, pain, anxiety and panic disorders, to more severe symptoms like disorders of perception, hallucinations, autonomic disturbances, seizures and delirium [25]. Minor alcohol withdrawal symptoms include tremulousness, diaphoresis, anxiety and palpitations. In more severe cases, patients develop seizures, alcoholic hallucinosis and delirium tremens [26]. In both situations, the administration of benzodiazepines is the treatment of choice and large doses may be needed until the desirable level of sedation and control of symptoms is achieved [24,27,28].

7. Thyroid storm

Thyroid storm, the most severe form of thyrotoxicosis, is considered a medical emergency. One in every six patients presenting with symptoms of thyrotoxicosis has a final diagnosis of thyroid storm. It has a high mortality rate, about 12-fold higher than in hospitalized patients with thyrotoxicosis without thyroid storm [29][30]. Other acute circumstances, such as infection, trauma, surgery, myocardial infarction, can precipitate this condition. Clinical manifestations may include hyperthermia, cardiac arrhythmias, gastrointestinal disorders and impaired mental status [30]. Other findings from the clinical presentation include ophthalmopathy, lid-lag, hand tremor and hyperreflexia [5]. Very low or undetectable TSH is the basic laboratory abnormality, but several diagnostic scores like the Burch-Wartofsky score or the Akamizu criteria have been proposed as adjuncts to the diagnosis [31]. Management of thyroid storm includes the administration of antithyroid drugs (propylthiouracil and methimazole), saturated solution of potassium iodide, hydrocortisone to decrease the conversion of T4 to T3 and beta-blockers for the relief of symptoms caused by the increased adrenergic activity. Supportive care with intravenous fluids, oxygen and cooling techniques is also essential, as well as treatment of precipitating factors [30].

8. Heat stroke

Heat-related illnesses are divided into three main

Table 5. *Differentiating features between sympathomimetic and antiholinergic toxidrome on physical examination.*

Organ system	Sympathomimetic	Anticholinergic
Ocular	Dilated and reactive pupils	Dilated and nonreactive pupils
Oral mucosa	Wet	Dry
Respiratory	Bronchodilator	Bronchodilator
Cardiovascular	Elevated blood pressure and tachycardia	Elevated blood pressure and tachycardia
Gastrointestinal	Normal bowel sounds	Decreased/absent bowel sounds
Genitourinary	Normal urination/occasionally urinary retention	Urinary retention
Skin	Diaphoretic	Red and dry
Neurologic	Psychomotor agitation	Hallucinations

categories. Minor heat illness (i.e. heat cramps, heat edema, heat syncope and prickly heat), heat exhaustion and heat stroke [2]. The clinical presentation depends on the body's ability to cope with heat. Heat stroke is the most severe illness and is defined on the basis of Tc > 40°C and neurologic dysfunction. There are two forms of heat stroke, the exertional and nonexertional. The exertional heat stroke is usually recognized in young individuals such as athletes, street workers and military recruits, who engage strenuous activity for a prolonged period of time in hot and dry environment [32,33]. In one study, the investigators observed that there was a 54% increase in risk for a work-related hyperthermia ED visit during an extreme heat event [34]. The classic nonexertional heat stroke usually affects the elderly or very young individuals and people with comorbidities during environmental heat waves. In most cases, it is attributed to the lack of adequate hydration and air conditioning. The presentation in these cases may be subtle and a high index of suspicion is required for the diagnosis [32]. During the next years, the number of ED visits, hospitalizations and deaths by heat stroke is expected to increase due to global warming [33]. There are several categories of people at greater risk of developing heat related illness (Table 6) [35].

When Tc is elevated, sweat production and cardiac output increase and heat dispersion through the skin is enhanced due to cutaneous vasodilation and visceral vasoconstriction. As these compensatory mechanisms fail and thermal homeostasis is disturbed, circulatory collapse emerges with catastrophic manifestations in multiple organs [4,35]. Typical symptoms include, except from hyperthermia and altered mental status, tachypnea, tachycardia, hypotension and hot, dry skin with absence of sweating. It can be complicated by myocardial ischemia, arrhythmias, rhabdomyolysis, acute kidney injury (AKI), acute hepatic failure, disseminated intravascular coagulation (DIC), gastrointestinal bleeding and electrolyte disturbances [2,4].

The diagnostic testing includes fingerstick glucose, complete blood count differential, coagulation testing, serum labs for the evaluation of electrolytes, renal and hepatic function, creatine kinase (CK) and cardiac biomarkers, urinalysis and electrocardiogram (ECG). Chest X-Ray, blood cultures, medication levels (if intoxication is suspected), neuroimaging, thyroid hormone levels etc. can be useful for the differential diagnosis [2,32]. Bedside ultrasound may assist in the estimation of ventricular filling and volume status [35].

The mainstay of treatment is rapid reduction of the Tc to about 39 °C because the duration of hyperthermia is related with the prognosis and the patient's outcome. However, the general measures, including insertion of two large bore intravenous catheters, rehydration, continuous cardiac and core temperature monitoring, administration of supplemental oxygen and early airway management, are of great importance. If the hemodynamic status is not supported adequately the risk of death and disabling neurological damage is higher [2,33,35]. Fluid replacement must be sufficient to restore hypotension and tissue perfusion. Due to lack of evidence for more specific recommendations (i.e. the type of fluid, the rate and volume of infusion), the therapeutic approach of fluid management in sepsis can be used as a guide to heatstroke because of the pathophysiological similarities between the two conditions [36]. The usual recommendation is that at least 30 ml/kg of IV crystalloids should be given within the first three hours. The

Table 6. *Risk factors for heat related illness.*

Non-modifiable risk factors	Modifiable risk factors
Age (the elderly and children)	Dehydration
Autonomic diseases with anhidrosis (Ross syndrome, Sjögren syndrome)	Prolonged activity in hot and humid environment
Spinal cord injuries	Occupational categories (military staff, athletes, outfield employees etc.)
Endocrine disorders (Diabetes, hyperthyroidism)	Abuse (alcohol, cocaine, amphetamine, opioids, etc.)
Neurological disorders (epilepsy)	Drugs (anticholinergics, beta-blockers, diuretics, neuroleptics, anesthetics)
Dermatological diseases (scleroderma, burns)	Infections
Hereditary disease (malignant hyperthermia)	Obesity

fluid resuscitation can be guided by the serum lactate level and the capillary refill time (CRT) as adjuncts to evaluate tissue perfusion, while dynamic measures including passive leg raising and cardiac output (CO) measurement can provide more accurate information [37]. The control of agitation is very important because the continuous muscle activity leads to further heat production, and it also increases the risk of rhabdomyolysis. So, the sedation of the agitated patient may be necessary regardless of whether the patient is intubated or not. Benzodiazepines are the first-line treatment of agitation, especially in cases of concurrent intoxication, as they act as muscle relaxants and because of their anti-seizure effect. Other options for sedation can be ketamine, propofol, opioids and dexmedetomidine. Benzodiazepines are also used for the suppression of shivering, which can impair temperature management if not controlled. If the patient is already intubated, paralysis with a non-depolarizing muscle-relaxant may be used for the treatment of shivering. Indications for intubation include the need for diagnostic procedures (i.e. lumbar puncture) in patients with altered mental status, status epilepticus, severe rigidity and the usual indications such as respiratory failure, refractory agitation etc. [32,33].

Cooling techniques

There are several methods of cooling. The choice of cooling technique depends on available resources and the cause of hyperthermia. The only absolute contraindication to cooling is a normal or low temperature. In cases of hyperthermia due to causes other than heat stroke, disease-specific treatment should not be delayed. Despite the method chosen, all clothes should be removed, and the patient should be totally exposed. The core temperature should be continuously monitored, and active cooling should be discontinued when Tc reaches 38-39 °C in order to avoid overshoot hypothermia. An esophageal or bladder probe is preferred but rectal temperature may be measured if the other options are unavailable. The standard equipment in an ER for the different techniques includes sheets/towels, a body bag, a cooling blanket, cold saline, ice packs, cool water bath, foley catheter, spray bottle and a fan [32,38].

1. Surface cooling

Surface cooling is the most important technique. An immersive ice-bath is considered to be the gold-standard for rapid cooling (achieving rates of cooling ~0.2 °C per minute). The technique includes filling a bath of water with ice until the water reaches a temperature of as low as 1-17 °C prior to the patient's immersion (true ice-water immersion). If the ice bath is not available, the patient can be placed in a body bag (or plastic sheets), which is filled with water and ice and then the body bag is closed up to the patient's neck. An alternative method of surface cooling is the evaporative cooling (cooling rates of ~0.1 °C per minute). The patient is sprayed with lukewarm water and then a fan is placed directly to the patient. A cooling blanket underneath the patient and ice packs on the groin, axilla and neck can be also used in order to maximize heat loss [32,39,40].

2. Internal cooling

Surface cooling is in general very effective but in patients presenting with shock can be delayed due to peripheral vasoconstriction. In such patients internal cooling using refrigerated crystalloid at 0-4 °C is beneficial. Although cooled fluids have not been studied in hyperthermia, their efficacy has been well established in other situations such as anesthesiology and neurocritical care [32]. The amount of IV fluids should be titrated with continuous core temperature measurement and patients with comorbidities, such as heart, liver or kidney failure, need special attention. Each 30 ml/kg bolus of 4 °C crystalloid is expected to reduce Tc by ~1,2-1,4 °C in hyperthermic patients, whereas the same dose of 20 °C crystalloid is expected to reduce Tc by 0,6-0,9 °C [41].

Other cooling methods are respiratory cooling (for short-term use) or more invasive methods like cold gastric, rectal or peritoneal lavage [35].

All patients should be hospitalized in order to be stabilized and observed for potential complications for at least 24 hours. Critically ill patients may worsen during cooling. In addition, skin is susceptible to damage from prolonged exposure to ice so it should be frequently checked during the cooling process. Close monitoring to avoid shivering that can impede cooling efforts, as well as hypothermia and its sequelae is crucial [42].

CONCLUSIONS

Hyperthermia may be difficult to diagnose in the emergency department setting. A great effort should be made so as all useful information is obtained regarding past medical and family history, drug history and the description of the event. A source of infection should be investigated in all cases with elevated body temperature but if the patient does not respond

to treatment with fluids and antibiotics emergency medicine physicians should seek for other causes of hyperthermia. The decrease of body core temperature is the most critical intervention in hyperthermic patients and along with disease-specific treatment, it should start as soon as possible in order to improve prognosis. Among cooling techniques, the immersive ice bath is considered the gold standard but other methods of surface cooling, which are simple to use, can be applied in the emergency department by an interprofessional team.

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REFERENCES

1. Balli S, Shumway KR, Sharan S. Physiology, Fever. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 [cited 2023 Jan 26]. Available from: http://www.ncbi.nlm.nih.gov/books/NBK562334/
2. Swaminathan A. Hyperthermia Syndromes [Internet]. REBEL EM - Emergency Medicine Blog. 2016 [cited 2022 Sep 13]. Available from: https://rebelem.com/hyperthermia-syndromes/
3. Hymczak H, Gołąb A, Mendrala K, Plicner D, Darocha T, Podsiadło P, et al. Core Temperature Measurement—Principles of Correct Measurement, Problems, and Complications. Int J Environ Res Public Health. 2021;18(20):10606.
4. Marchand M, Gin K. The Cardiovascular System in Heat Stroke. CJC Open. 2022;4(2):158–63.
5. Boushra M. Evaluation and Treatment of Elevated Temperature in the Emergency Department [Internet]. Trauma and Emergency Surgery - The Role of Damage Control Surgery. IntechOpen; 2020 [cited 2022 Sep 13]. Available from: www.intechopen.com/chapters/undefined/state.item.id
6. Simon LV, Hashmi MF, Callahan AL. Neuroleptic Malignant Syndrome. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 [cited 2022 Sep 20]. Available from: http://www.ncbi.nlm.nih.gov/books/NBK482282/
7. Ngo V, Guerrero A, Lanum D, Burgett-Moreno M, Fenati G, Barr S, et al. Emergent Treatment of Neuroleptic Malignant Syndrome Induced by Antipsychotic Monotherapy Using Dantrolene. Clin Pract Cases Emerg Med. 2019;3(1):16–23.
8. Spadaro A, Scott KR, Koyfman A, Long B. High risk and low prevalence diseases: Serotonin syndrome. Am J Emerg Med. 2022;61:90–7.
9. Boyer EW, Shannon M. The serotonin syndrome. N Engl J Med. 2005;352(11):1112–20.
10. Chiew AL, Buckley NA. The serotonin toxidrome: shortfalls of current diagnostic criteria for related syndromes. Clin Toxicol Phila Pa. 2022;60(2):143–58.
11. Graudins A, Stearman A, Chan B. Treatment of the serotonin syndrome with cyproheptadine. J Emerg Med. 1998;16(4):615–9.
12. Nelson TE. Heat production during anesthetic-induced malignant hyperthermia. Biosci Rep. 2001;21(2):169–179.
13. Wappler F. Malignant hyperthermia. Eur J Anaesthesiol. 2001;18(10):632–52.
14. Larach MG, Gronert GA, Allen GC, Brandom BW, Lehman EB. Clinical presentation, treatment, and complications of malignant hyperthermia in North America from 1987 to 2006. Anesth Analg. 2010;110(2):498–507.
15. Larach MG, Localio AR, Allen GC, Denborough MA, Ellis FR, Gronert GA, et al. A clinical grading scale to predict malignant hyperthermia susceptibility. Anesthesiology. 1994\;80(4):771–9.
16. Hopkins PM, Girard T, Dalay S, Jenkins B, Thacker A, Patteril M, et al. Malignant hyperthermia 2020. Anaesthesia. 2021;76(5):655–64.
17. Zvaritch E, Gillies R, Kraeva N, Richer M, Jungbluth H, Riazi S. Fatal awake malignant hyperthermia episodes in a family with malignant hyperthermia susceptibility: a case series. Can J Anesth Can Anesth. 2019;66(5):540–5.
18. Timmins MA, Rosenberg H, Larach MG, Sterling C, Kraeva N, Riazi S. Malignant hyperthermia testing in probands without adverse anesthetic reaction. Anesthesiology. 2015;123(3):548–56.
19. O'Malley GF. Emergency Department Management of the Salicylate-Poisoned Patient. Emerg Med Clin North Am. 2007;25(2):333–46.
20. Salicylate Intoxication | NEJM [Internet]. [cited 2023 Feb 7]. Available from: https://www.nejm.org/doi/10.1056/NEJM197305242882107?url_ver=Z39.88-2003&rfr_id=ori:rid:crossref.org&rfr_dat=cr_pub%20%200pubmed
21. Bongers KS, Salahudeen MS, Peterson GM. Drug-associated non-pyrogenic hyperthermia: a narrative review. Eur J Clin Pharmacol. 2020;76(1):9–16.
22. Dawson AH, Buckley NA. Pharmacological management of anticholinergic delirium - theory, evidence and practice. Br J Clin Pharmacol. 2016;81(3):516–24.
23. Hayes BD, Martinez JP, Barrueto F. Drug-Induced Hyperthermic Syndromes: Part I. Hyperthermia in Overdose. Emerg Med Clin North Am. 2013;31(4):1019–33.
24. Kosten TR, O'Connor PG. Management of Drug and Alcohol Withdrawal. N Engl J Med. 2003;348(18):1786–1795.
25. Soyka M. Treatment of Benzodiazepine Dependence. N Engl J Med. 2017;376(12):1147–57.
26. Wijdicks EFM. The Discovery of Acute Alcohol Withdrawal as a Cause of Delirium. Neurocrit Care. 2022;37(3):806–809.
27. Amato L, Minozzi S, Vecchi S, Davoli M. Benzodiazepines for alcohol withdrawal. Cochrane Database Syst Rev [Internet]. 2010 [cited 2023 Feb 7];(3). Available from: https://www.cochranelibrary.com/cdsr/doi/10.1002/14651858.CD005063.pub3/full
28. DeBellis R, Smith BS, Choi S, Malloy M. Management of Delirium Tremens. J Intensive Care Med. 2005;20(3):164–173.

29. Galindo RJ, Hurtado CR, Pasquel FJ, García Tome R, Peng L, Umpierrez GE. National Trends in Incidence, Mortality, and Clinical Outcomes of Patients Hospitalized for Thyrotoxicosis With and Without Thyroid Storm in the United States, 2004–2013. Thyroid. 2019;29(1):36–43.
30. Leung AM. Thyroid Emergencies. J Infus Nurs Off Publ Infus Nurses Soc. 2016;39(5):281–6.
31. Akamizu T, Satoh T, Isozaki O, Suzuki A, Wakino S, Iburi T, et al. Diagnostic Criteria, Clinical Features, and Incidence of Thyroid Storm Based on Nationwide Surveys. Thyroid. 2012l;22(7):661–79.
32. Hyperthermia & heat stroke [Internet]. EMCrit Project. [cited 2022 Sep 13]. Available from: https://emcrit.org/ibcc/hyperthermia/
33. Heat Stroke: Practice Essentials, Pathophysiology, Etiology [Internet]. [cited 2022 Sep 13]. Available from: https://emedicine.medscape.com/article/166320-overview#a1
34. Shire J, Vaidyanathan A, Lackovic M, Bunn T. Association Between Work-Related Hyperthermia Emergency Department Visits and Ambient Heat in Five Southeastern States, 2010–2012—A Case-Crossover Study. GeoHealth. 2020;4(8):e2019GH000241.
35. Savioli G, Zanza C, Longhitano Y, Nardone A, Varesi A, Ceresa IF, et al. Heat-Related Illness in Emergency and Critical Care: Recommendations for Recognition and Management with Medico-Legal Considerations. Biomedicines. 2022;10(10):2542.
36. Bouchama A, Dehbi M, Chaves-Carballo E. Cooling and hemodynamic management in heatstroke: practical recommendations. Crit Care. 2007 12;11(3):R54.
37. Evans L, Rhodes A, Alhazzani W, Antonelli M, Coopersmith CM, French C, et al. Surviving sepsis campaign: international guidelines for management of sepsis and septic shock 2021. Intensive Care Med [Internet]. 2021 [cited 2023 Apr 18];47(11). Available from: https://link.springer.com/epdf/10.1007/s00134-021-06506-y
38. Wasserman DD, Creech JA, Healy M. Cooling Techniques For Hyperthermia. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 [cited 2022 Sep 13]. Available from: http://www.ncbi.nlm.nih.gov/books/NBK459311/
39. Wang AZ, Lupov IP, Sloan BK. A Novel Technique for Ice Water Immersion in Severe Drug-Induced Hyperthermia in the Emergency Department. J Emerg Med. 2019;57(5):713–5.
40. First aid cooling techniques for heat stroke and exertional hyperthermia: A systematic review and meta-analysis - Resuscitation [Internet]. [cited 2023 Jan 26]. Available from: https://www.resuscitationjournal.com/article/S0300-9572(20)30028-9/fulltext
41. Blumenberg A. Dosing Heat: Expected Core Temperature Change with Warmed or Cooled Intravenous Fluids. Ther Hypothermia Temp Manag. 2021;11(4):223–9.
42. Wasserman DD, Thurman J, Healy M. EMS Methods To Cool A Patient In The Field. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 [cited 2022 Sep 13]. Available from: http://www.ncbi.nlm.nih.gov/books/NBK459303/
43. Differential diagnosis of hyperthermia* - UpToDate [Internet]. [cited 2022 Sep 13]. Available from: https://www.uptodate.com/contents/image?imageKey=EM%2F53571

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Adrenal Insufficiency: A review

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Abstract

Adrenal insufficiency (AI) is defined by a complete or partial lack of adrenal glucocorticoids with or without concomitant lack of mineralocorticoids and adrenal androgens. Primary adrenal insufficiency is an uncommon and serious disease of loss of function of the adrenal glands. Secondary AI (SAI) is more common and is brought on by conditions that affect the pituitary, whereas tertiary AI (TAI) is brought on by conditions that affect the hypothalamus. TAI is the most common kind due to withdrawal of exogenous glucocorticoids. The lack of cortisol, adrenal androgen precursors, and aldosterone (particularly in PAI) are linked to the non-specific, frequently missed, or incorrectly diagnosed symptoms of AI. The assessment of adrenal corticosteroid hormones, their regulatory peptide hormones, and stimulation tests serve as the foundation for diagnosis. Establishing a hormone-replacement regimen that closely resembles the body's natural diurnal cortisol secretion rhythm and is adapted to the patient's daily needs is the main objective of therapy.

Key words: *Adrenal insufficiency; addison disease; ηypothalamic-pituitary-adrenal axis*

INTRODUCTION

Adrenal hypofunction, which results in insufficient synthesis of glucocorticoids, particularly cortisol, characterises the endocrine condition known as adrenal insufficiency (AI). According to the underlying aetiology, AI can be primary, secondary, or tertiary. Hence, the synthesis of mineralocorticoids and adrenal androgens may also be impacted [1]. When the adrenal glands are damaged or destroyed, it results in primary adrenal insufficiency (PAI), which is a direct failure of the adrenal glands. The most common cause of PAI, particularly in high-income countries, is autoimmune adrenalitis, also known as Addison disease. In contrast, the proportion of patients with PAI whose AI is caused by infectious diseases, such as tuberculosis or human immunodeficiency virus (HIV), is high in nations with high prevalence of these infections [2–4]. The condition known as secondary adrenal insufficiency (SAI) results from conditions affecting the pituitary or the hypothalamus, such as tumours of the pituitary and/or hypothalamus and the treatment options (surgery and/or radiation), hypophysitis, or granulomatous infiltration. Tertiary adrenal insufficiency (TAI) is characterised by hypothalamic anomalies or dysfunction, which results in decreased corticotropin-releasing hormone (CRH) output. Similar to SAI, the subsequent absence of adrenocorticotropic hormone (ACTH) stimulation results in decreased cortisol and dehydroepiandrosterone (DHEA) release. As a result, TAI is frequently referred to as SAI without any further distinction between the two terms. TAI often develops from the persistent use of supraphysiological amounts of exogenous glucocorticoids, which suppresses the hypothalamic-pituitary-adrenal (HPA) axis [5].

The clinical manifestations of AI that frequently occur gradually over a lengthy period of time are associated with the corresponding hormone shortage. Fatigue, hypotension, and weight loss are three major symptoms especially linked to cortisol deficit; nevertheless, because they are vague, they frequently cause diagnostic delays and incorrect diagnoses [6]. Particularly, unexplained hyponatraemia and hyperpigmentation or hair loss in

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the pubis and axillae may point to AI. Adrenal crisis is a severe emergency that can also appear as AI. Patients with unknown AI and those receiving established replacement treatment run the danger of suffering a life-threatening adrenal crisis due to the critical protective role of cortisol under stressful circumstances [7]. Hence, one of the main objectives of long-term management is to prevent adrenal crises.

Iatrogenic causes of AI have significant implications for several medical specialties. Opioids and glucocorticoids, which are known to impede the HPA axis's function, are commonly utilized to treat a variety of illnesses. The incidence of AI has further increased due to the more widespread use of immune checkpoint inhibitors (ICIs), which are linked to autoimmune hypophysitis and infrequently to autoimmune adrenalitis.

Hypothalamus-Pituitary Axis

Adrenal glands consist of adrenal cortex and medulla. Zonas fasciculata, glomerulosa, and reticularis make up the three zones of the cortex. Cortisol is produced by the zona fasciculata, mineralocorticoids (aldosterone) are produced by the zona glomerulosa, and sex hormones are produced by the zona reticularis. Catecholamines are produced by the adrenal medulla. Adrenal glands use cholesterol as a starting material to create steroid hormones.

The hypothalamus-pituitary axis regulates the adrenal cortex's ability to secrete cortisol. The paraventricular nucleus (PVN) of the hypothalamus secretes both arginine vasopressin and CRH. PVN produces and releases CRH and vasopressin in response to stress inputs from other brain nuclei as well as circadian input from the hypothalamic suprachiasmatic nucleus. Portal vein CRH stimulates pituitary corticotroph cells to secrete ACTH. Cortisol, DHEA, and, to a lesser extent, aldosterone secretion are stimulated by ACTH when it binds to certain receptors in the adrenal cortex (melanocortin-2-receptor-MC2R) [8]. A negative feedback loop regulates cortisol secretion by binding to particular receptors in the brain and the pituitary and preventing ACTH secretion [9]. While ACTH release from pituitary corticotroph cells precedes cortisol peaks, the circadian rhythm of cortisol secretion is characterized by increased levels in the morning (peak after awakening) and nadir levels in the evening (before bedtime) [10]. A negative brief feedback loop between cortisol and ACTH controls the release of cortisol as well (ultradian rhythmicity) [11].

Cortisol acts by binding to particular cytoplasmatic receptors, and this complex then moves to the nucleus, where it binds to particular glucocorticoid and mineralocorticoid response elements. This process results in the transcription or repression of particular genes, which is necessary for cortisol's physiological function. By its interaction with particular glucocorticoid receptors and mineralocorticoid receptors, cortisol plays a role in a number of physiological processes including metabolism, stress response, reproduction, immunity, and cognition [12,13]. Plasma proteins bind to cortisol once it has been secreted into the bloodstream, keeping it inactive. 80–90% of cortisol is bound to corticosteroid-binding globulin (CBG), the most significant binding protein. Many variables, like body temperature or inflammation, have an impact on CBG levels. A decrease in CBG affinity and an increase in free cortisol are observed in a variety of pathologic conditions, such as fever or inflammation [14,15].

The mineralocorticoids are secreted by the zona glomerulosa of the adrenal glands (aldosterone). The renin-angiotensin system and potassium are the primary regulators of aldosterone secretion. The renin-angiotensin system controls electrolyte balance, blood volume, and blood pressure. When sodium concentration is low or when renal perfusion pressure is reduced, the kidneys produce renin. Angiotensinogen is processed by renin into angiotensin I, which is then transformed into angiotensin II by the angiotensin-converting enzyme. Angiotensin II increases the release of aldosterone from the adrenal cortex by binding to angiotensin receptors. Aldosterone production is stimulated by elevated serum potassium levels. Moreover, ACTH stimulates aldosterone secretion, whereas glucocorticoids inhibit it [16–18].

The C19 steroids produced by the adrenal glands include DHEA and DHEAS, which have received the most attention as potential indicators of excessive androgen production in the adrenal glands. DHEAS is the most abundant C19 steroid secreted by the adrenal glands, according to recent measurements of steroids using liquid chromatography/tandem mass spectrometry in samples taken directly from the adrenal veins. However, it is shown that the adrenal gland is the source of other C19 steroids. ACTH seems to be the main mediator, even if the processes that control the synthesis of adrenal C19 steroid have not yet been fully understood. Contrary to the testis, which is a highly effective androgen generator, the adrenal gland only contributes minimally to the production of active androgen in males. However, the adrenal gland is a significant source of androgen

and androgen precursors in women and pre-pubertal children, which have physiologic and pathologic functions. The adrenal glands' participation in the synthesis of pathologic androgen is crucial in a number of conditions. Future research is required to determine the mechanisms governing adrenal C19 steroidogenesis control and dysregulation in both normal and pathologic production.

A. EPIDEMIOLOGY AND PATHOGENESIS

The types of adrenal insufficiency are depicted in Figure 1.

Primary Adrenal Insufficiency

The prevalence and range of underlying causes of PAI have varied throughout the past century, according to the epidemiology of the condition. With an incidence of 10–20 cases per 100,000 people, PAI is a rare condition with several possible causes. In industrialized countries, Addison's disease accounts for about 90% of non-congenital adrenal hyperplasia (CAH) cases, but primary adrenal insufficiency from infections (tuberculosis, Cytomegalovirus, HIV) is more prevalent in developing countries [4,19–22]. CAH is a typical contributor to primary adrenal insufficiency [3,23]. The prevalence of Addison's disease varies by region, from 1.4 cases per 100,000 people in South Africa to 9–22 cases per 100,000 people in Europe [24–26]. Addison's disease can develop at any age, but its average onset age is between 20 and 50 years. It also occurs more frequently in people with other autoimmune disorders, such as those with type 1 diabetes mellitus, compared to the general population [27]. The most prevalent inherited form of PAI is CAH, which is brought on most commonly by a 21-hydroxylase deficiency. One in fifty patients with CAH, an autosomal recessive condition, have a CYP21A2 mutation (encoding 21- hydroxylase). Between 0.5 and 1 cases of classic CAH are found in every 10,000 people, depending on the population. Adrenoleukodystrophy, adrenal hypoplasia congenita, and autoimmune polyglandular syndrome (APS) type 1 are additional, less common hereditary causes of PAI [28]. Because CAH lacks 21-hydroxylase, there is less cortisol produced, which in turn causes the pituitary to secrete more ACTH. The reactions catalysed by 21-hydroxylase are the conversion of progesterone in 11-deoxycorticosterone and of 17-OH progesterone in deoxycortisol. Adrenocortical hyperplasia and the release of steroidogenic products upstream of the enzymatic block, such as adrenal androgens, are induced by increasing ACTH. The underlying mutation and the remaining enzyme activity determine the severity of CAH. Neonates have low levels of cortisol and aldosterone when the enzyme activity is less than 2%. In addition to having defective aldosterone synthesis, the severe type of CAH, known as salt-wasting syndrome, also causes virilization of the external genitalia in female infants as a result of excessive adrenal androgen production [29]. There are symptoms of virilization, premature pseudopuberty, or growth arrest in less severe mutations. Milder forms of CAH, often known as "non-classic CAH" or "late-onset" CAH, are more

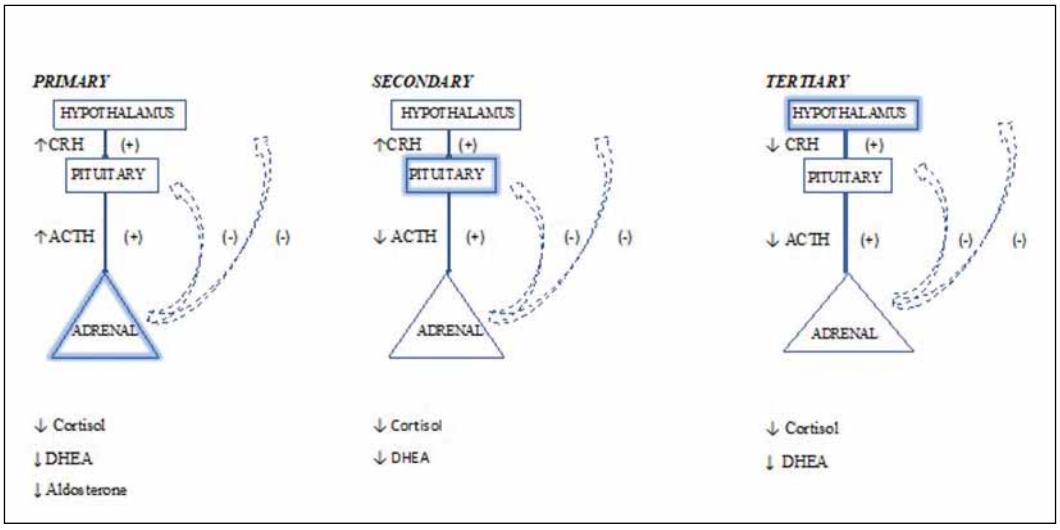

Figure 1. Types of adrenal insufficiency.

prevalent. Aldosterone synthesis is normal, cortisol insufficiency may be slight or nonexistent, and adrenal androgens are overproduced in this variant of CAH [30]. A number of autoimmune comorbidities, including autoimmune thyroid disease (which affects 40% of patients), premature ovarian failure (5–16%), type 1 diabetes mellitus (11%), pernicious anemia (10%), vitiligo (6%), and celiac disease (2%), coexist with Addison disease [31].

Autoantibodies against 21-hydroxylase are typically detected in people with autoimmune Addison disease. It appears that CD4+ and CD8+ T lymphocytes that are active against 21-hydroxylase are responsible for adrenal cortex destruction. Antibodies to 21-hydroxylase can be found with great specificity and sensitivity, and their finding can occur several years before the manifestation of the disease. Additional research has linked specific major histocompatibility complex (MHC) genotypes, including DR3-DQ2 and DR4-DQ8, to Addison disease. The genetic causes of Addison disease make it another inherited illness [32–36]. The production of mineralocorticoids is impaired in primary adrenal illness, which also involves the zona glomerulosa. Lack of aldosterone causes salt loss, decreased fluid volume, and hypotension, which affects the electrolyte balance. The darkening of the skin (bronzing), which is particularly pronounced in sun-exposed areas and over pressure points like the elbows, scars, knees, and the oral mucosa, is a defining symptom of people with Addison disease. Hyperpigmentation of the skin and mucous membrane can precede over other signs by months to years. Lack of cortisol feedback causes increased release of pro-opiomelanocortin (POMC), as well as peptides generated from it, such as ACTH and melanocyte stimulating hormone (MSH). Increased MSH stimulates melanocortin receptor 1 to cause hyperpigmentation [37] (Table 1).

Secondary Adrenal Insufficiency

It has been suggested that SAI is more widespread than PAI, with a prevalence of up to 42 cases per 100,000 people. Only rarely can SAI have a genetic basis (for instance, mutations of the transcription factor TPIT).

Table 1. *Causes of primary adrenal insufficiency*

Aetiology	Pathogenesis	Diagnostic tools
Genetic	Congenital adrenal hyperplasia, congenital lipoid adrenal hyperplasia, adrenoleukodystrophy (X-linked), adrenal hypoplasia congenita, autoimmune polyglandular syndrome type 1	Sequence of relevant gene
Autoimmune	T and B cell autoimmunity against adrenocortical cells	21- Hydroxylase autoantibodies
Infiltrative	Amyloidosis Haemochromatosis Histiocytosis	Adrenal CT, subcutaneous fat biopsy Ferritin, HFE sequencing Adrenal imaging
Tumour	Primary tumour (bilateral), metastasis (bilateral), adrenal lymphoma (bilateral)	Adrenal CT
Infection	Mycobacteria, bacteria (e.g. Neisseria meningitidis, Haemophilus influenzae, Pseudomonas aeruginosa), viruses (e.g. human immunodeficiency virus, herpes simplex, cytomegalovirus) or fungi (e.g. Pneumocystis jirovecii)	Culture, QuantiFERON test, PCR, adrenal CT
Surgery	Bilateral adrenalectomy	Patient history
Bleeding	Anti- phospholipid syndrome, anticoagulant therapy, disseminated intravascular coagulation	Adrenal CT, phospholipid autoantibodies
Medication	Enzyme inhibition (ketoconazole, fluconazole, itraconazole, etomidate, aminoglutethimide, metyrapone, trilostane, osilodrostat); adrenolytic effect and increased cortisol metabolism (mitotane); inflammation (checkpoint inhibitors)	Medication and patient history

Usually, it is reported in people with disorders that affect the pituitary, such as tumours, autoimmune hypophysitis, trauma, or after radiotherapy of intracranial tumors [38,39] (Table 2).

Because of the pituitary gland's mass effect from the tumour, SAI will manifest in 10–62% of individuals with pituitary adenoma; the risk rises with pituitary surgery or radiotherapy. Pituitary insufficiency after radiotherapy is usually seen many years later and is dose dependent [40]. Moreover, SAI has been documented in 26–57% of patients with hypophysitis, compared to 23% of individuals with any type of pituitary stalk lesion [41–43].

In patients receiving immune checkpoint inhibitor therapy for various forms of cancer, hypophysitis is a relatively frequent finding (13%) [44].

Chronic opiate use, even with low doses of morphine, is another factor that contributes to secondary adrenal insufficiency that is iatrogenic and results from pituitary insufficiency. It is a pathological syndrome that is underestimated and not well understood. It may be caused by both an opioid's detrimental impact on the production of adrenal glucocorticoids and a disruption of the circadian rhythm of glucocorticoids. Long-term opioid usage can also damage the gonadotroph axis and cause secondary hypogonadism [45,46].

Tertiary Adrenal Insufficiency

Endogenous causes such as tumours, radiation, or inflammatory processes that impact the hypothalamus may contribute to tertiary AI. Chronic exogenous glucocorticoid injection is the main cause of TAI. In the majority of individuals, glucocorticoid-induced TAI, also known as iatrogenic or exogenous AI, is a transient condition. The risk of developing TAI varies depending on the dose and the route of glucocorticoid administration. Intranasal administration is stated to carry a 4.2% risk of TAI, inhalation is claimed to carry a 20% risk, oral administration a 49% risk, and intra-articular delivery a 52% risk. With short-term, low-dose glucocorticoid usage, the risk of AI is lowest, while with long-term, high-dose glucocorticoid use, the risk is highest. Yet, the relationship between TAI and glucocorticoid seems to be multifactorial, suggesting that other factors may also affect a person's vulnerability [47]. Every patient receiving glucocorticoid medication must be thought of as having a TAI risk; as a result, the dosage must be carefully tapered and the patient must receive counseling. This advice is further backed by the higher death rate seen in the first three months after stopping glucocorticoid medication [48]. Restoration of HPA axis function is usually observed after 24 months of cessation of glu-

Table 2. *Causes of secondary adrenal insufficiency.*

Aetiology	Pathogenesis	Clinical manifestations and diagnostic tools
Genetic	Mutations in transcription factors, hormone and hormone receptor genes	Sequence of relevant gene
Autoimmune	T and B cell autoimmunity against pituitary cells (hypophysitis), IgG4- related hypophysitis	Pituitary MRI, confirmation of subtype by transsphenoidal biopsy, screen for hormone deficiencies
Tumour	Pituitary adenoma, craniopharyngioma, metastasis (carcinomas of breast, lung, colon, prostate), chondroma, chordoma, germinoma, suprasellar meningioma, astrocytoma of the optic nerve, ependymoma, pituitary carcinoma, lymphoma	Pituitary MRI, screen for hormone deficiencies
Infiltrative	Sarcoidosis, histiocytosis, haemochromatosis, amyloidosis	Pituitary MRI, specific diagnostic tests
Iatrogenic	Pituitary surgery, radiotherapy	Pituitary MRI, screen for hormone deficiencies
Traumatic and vascular	Traumatic brain injury, postpartum apoplexy (Sheehan syndrome), tumour apoplexy, subarachnoidal bleeding, aneurysm, snake bite	Severe headache, visual disturbances, pituitary MRI, screen for hormone deficiencies
Infectious	Tuberculosis, HIV infection, meningitis, actinomycosis, syphilis	Pituitary MRI, specific diagnostic tests
Medication	Checkpoint inhibitors, interferon- α, opioids	Patient history

cocorticoid treatment, however, some individuals may experience a longer recovery period of up to 4 years [49,50] (Table 3).

Adrenomedullary function in AI

Catecholamine production under physiological settings and in response to stress is impaired in patients with primary and secondary adrenal insufficiency [51,52]. In particular, norepinephrine levels rise while epinephrine output falls. The root reason is not entirely known. It is well known that high local glucocorticoid concentrations from the adrenal cortex via a rich blood supply are necessary for the action of the enzyme PNMTase, which catalyses the conversion of norepinephrine to epinephrine [53]. In patients with adrenal insufficiency, it appears that glucocorticoid replacement may not achieve the requisite glucocorticoid levels for enzyme activation. It is unclear what clinical significance low epinephrine levels have. Apparently, it may be responsible for the diminished well-being, the cognitive impairment, and the diminished glucose counter-regulatory response to hypoglycaemia [54,55].

Adrenal androgens

The zona reticularis of the adrenal glands produces DHEA. Due to the loss of the zona reticularis in the first case and the impairment of ACTH production in the second, it is seen that primary adrenal insufficiency (apart from CAH) and secondary adrenal insufficiency have impaired DHEA secretion. Symptoms of DHEA deficiency are observed in women, given that testosterone production in men is preserved. Adrenal androgens' physiological function is not entirely known. The DHEA effects are direct and indirect. Indirect actions result in the conversion of these adrenal androgen precursors into active androgens or estrogens in the target organs such as gonads. The direct actions are related to the immunomodulatory action and to the neuroprotective effect in the brain and this may be one of the reasons for the impairment of well-being in patients with adrenal insufficiency [56].

B. CLINICAL MANIFESTATIONS

Adrenocortical hormones are vital to the organism and their deficiency causes a host of symptoms affecting almost all systems. Symptoms vary depending on the cause of the adrenal insufficiency, as shown in Table 4.

C. DIAGNOSIS

Primary Adrenal Insufficiency

The importance of raising awareness of PAI is highlighted by the fact that the majority of patients expe-

Table 3. *Drugs inducing adrenal insufficiency.*

Compounds	Mechanism
Supraphysiological treatment with glucocorticoid hormones	Suppression of the HPA axis through negative feedback on the hypothalamus and pituitary and induction of tertiary adrenal insufficiency in case of long- term treatment
Immune checkpoint inhibitors	Hypophysitis due to lymphocytic pituitary infiltration of lymphocytes leading to ACTH deficiency; less common, adrenalitis and direct impairment of adrenal cortex function (new immune checkpoint inhibitors seem to cause hypophysitis less frequently)
IFNα	Induction of hypophysitis
Rifampicin, phenytoin, troglitazone, phenobarbital	Increased cortisol metabolism by induction of CYP3A4
Mitotane	Destruction of adrenocortical tissue (adrenolytic effect); strong induction of the cortisol- metabolizing enzyme CYP3A4
Adrenostatic agents	Inhibition of corticosteroid synthesis by inhibition of steroidogenic enzymes, mainly 11β- hydroxylase (CYP11B1) and aldosterone synthase (CYP11B2), side- chain cleavage enzyme (CYP11A1)
Mifepristone	Glucocorticoid receptor antagonist; adrenal insufficiency occurs in overdosing
Opioids	Inhibition of the HPA axis by blocking release of hypothalamic corticotropin- releasing hormone; also affect adrenal steroid production

Table 4. *Clinical manifestations of adrenal insufficiency.*

System	Clinical manifestations
Central nervous system	Anorexia[1]
	Weight loss[1]
	Nausea[1]
	Salt craving[2]
	Dizziness[2]
Cardiovascular system	Hypotension and/or dehydration
Haematological system	Anaemia[1]
	Lymphocytosis
	Eosinophilia[1]
Blood electrolytes	Hyponatraemia[1,2]
	Hyperkalaemia[2]
	Hypercalcaemia[1]
Neuropsychiatric system	Depression[1,3]
	Fatigue[1]
	Reduced libido[3]
Gastrointestinal system	Diarrhea
	Vomiting[1]
	Abdominal pain[1]
Dermatological system	Dry skin[3]
	Hyperpigmentation (PAI)
	Hypopigmentation (SAI)
	Loss of pubic/axillary hair[3]
Musculoskeletal system	Myalgia[1]
	Joint pain
	Weakness[1]

[1]Symptoms specific to glucocorticoid deficiency
[2]Symptoms specific to mineralocorticoid deficiency
[3]Symptoms specific to adrenal androgen deficiency

rience non-specific symptoms such as fatigue, poor health, postural dizziness, nausea, and weight loss for years before being diagnosed, and that these symptoms are frequently attributed to other causes. Diagnostic tests should never be run (or the results of tests should never be awaited before starting treatment for suspected acute manifestations of AI).

Typical symptom of primary adrenal insufficiency is changes in skin pigmentation. Skin hyperpigmentation is a characteristic finding of primary adrenal insufficiency, but the degree of hyperpigmentation varies among patients. Another cardinal finding in these patients is low blood pressure and orthostatic hypotension, because of aldosterone deficiency. Common laboratory findings are hyponatraemia, hyperkalaemia and hypoglycaemia. DHEA deficiency is accompanied by loss of pubic and axillary hairs and decreased libido in women, while men do not manifest similar disorders. Adrenal crisis could be the first presentation of the disease.

A coupled assessment of blood cortisol and plasma ACTH serves as the diagnostic test for suspected PAI. When combined with elevated ACTH levels (twice the upper normal limit), a morning serum cortisol level of less than 5 g/dl indicates PAI. It is advised to confirm the reference ranges with the appropriate laboratory because cortisol concentrations fluctuate depending on the assay utilized [57]. Due to improved test specificity, mass spectrometry analyses have been demonstrated to detect lower cortisol concentrations than immunoassays [58]. In patients with cortisol values >5 μg/dl, confirmation of the diagnosis is made using ACTH stimulation test (Synacthen test or corticotropin-stimulation test, 250-μg dose of synthetic ACTH 1–24). A peak serum cortisol concentration of <16 μg/dl min 30 after ACTH, or a peak <18 μg/dl 60 min after ACTH confirms the diagnosis [59]. To determine the presence of mineralocorticoid deficit, which is characterised by low or low-to-normal aldosterone in the presence of elevated levels of renin, combined plasma renin and aldosterone levels should be tested. The aetiology should be looked into, once PAI has been diagnosed. Patients should be examined for the presence of 21-hydroxylase autoantibodies in the serum in areas with a high frequency of Addison disease. Patients who have autoantibody tests that are positive should be checked for concurrent autoimmune comorbidities. However, testing negative for 21-hydroxylase autoantibodies does not rule out autoimmune PAI. To rule out inflammatory processes, adrenal gland destruction due to haemorfrhage or infiltration, such as bilateral extra-adrenal cancer metastases, in patients with negative autoantibody tests, CT imaging of the adrenal region is recommended [60]. Adrenoleukodystrophy should be ruled out in men with negative 21-hydroxylase autoantibodies by checking the serum levels of very long chain fatty acids.

Secondary Adrenal Insufficiency

In secondary adrenal insufficiency, in addition to symptoms of glucocorticoid deficiency, there are also symptoms from the absence of other pituitary hormones (hypothyroidism, hypogonadism) or symptoms due to pressure of the tumor in the adjacent structures (headache, visual field disturbances).

In individuals with known or suspected pituitary illness, determining the HPA axis' stress reactivity is crucial. With structural pituitary pathology, such as tumours or injuries, the corticotropic axis is often one of the last anatomical units to be destroyed after the thyrotropic axis, albeit autoimmune hypophysitis can result in solitary ACTH insufficiency. Hyponatraemia with a history of stroke or traumatic brain injury and symptoms of attention and cognitive deficits, nausea, vomiting, disorientation, headache, somnolence, and seizures should warrant additional testing for a possible SAI [61].

Although serum cortisol > 16 μg/dl precludes secondary adrenal insufficiency, a morning serum cortisol 3,6 g/dl and a low or low-normal ACTH level are indicative of the condition [61]. The insulin tolerance test is the gold standard test to assess the integrity of the hypothalamus-pituitary axis. A severe stress reaction and the release of hormones that counteract it, including cortisol and growth hormone, are brought on by hypoglycemia. The test is not safe for people who have a history of cardiovascular illness, stroke, or epilepsy due to the extreme hypoglycaemia that is induced. The test is carried out in specialist facilities as a result. The ACTH test is the most often utilized test since it is reliable and secure. Peak serum cortisol levels above 18 g/dl are regarded as normal. The ACTH test should be administered six weeks after pituitary surgery when it's necessary to assess cortisol reserve following transsphenoidal surgery for a pituitary adenoma, because, the adrenal cortisol response to an ACTH test seems to be normal for a brief period of time. Synthetic CRH testing is a great way to assess hypothalamic function, but it doesn't seem to be any better than ACTH stimulation testing, and some facilities have restrictions on its use. Metyrapone and the glucagon test are additional tests for cortisol reserve evaluation, but they are rarely used. Measuring basal salivary cortisol and salivary cortisone is a potential technique for the diagnosis and screening of secondary adrenal insufficiency due to the simplicity of the procedure and independence from binding proteins like albumin or CBG.

Under stressful physical or psychological settings, patients with undiagnosed adrenal insufficiency are susceptible to developing a life-threatening adrenal crisis. For this reason, it's critical to include adrenal deficiency in the differential diagnosis for all patients who have unexplained hyponatraemia, hypotension, vomiting, or diarrhea as well as those who present with atypical symptoms. When there is a clinical suspicion of Addison disease, patients with other autoimmune disorders such as type 1 diabetes, hypothyroidism, premature ovarian failure, pernicious anemia, etc. should be examined [61–65].

D. MANAGEMENT

According to the European Society of Endocrinology a successful treatment aim to achieve an optimal glucocorticoid dosing regimen in order to avoid complications from overtreatment such as metabolic syndrome, cardiovascular disease and osteoporosis, but also to ensure a good quality of life and daily performance and to prevent adrenal crisis [66].

Glucocorticoid replacement

The purpose of hormone replacement is to mimic the diurnal sleep pattern of cortisol secretion as closely as possible. Until now there is no therapeutic preparation that mimics the cortisol secretion profile. The optimal glucocorticoid dosing is based primarily on the patient's symptoms, since there are no biological markers indicating cortisol sufficiency or not. Cortisol measurement is not a reliable indicator of ideal glucocorticoid replacement, specifically when prednisolone is used. It is recommended that titration of the glucocorticoid dose should be based on patient symptoms of cortisol deficiency and excess. Usually patients with insufficient glucocorticoid replacement complain of fatigue, weight loss and nausea, while glucocorticoid overtreatment induce a picture of iatrogenic Cushing with increased abdominal fat, thin skin, easy bruising, hypertension and type 2 diabetes mellitus. Usually patients with PAI require greater glucocorticoid replacement compared to patients with SAI, owing to residual ACTH secretion.

The pharmaceutical preparation indicated for the treatment of adrenal insufficiency is hydrocortisone. Hydrocortisone is administered 2 or 3 times daily, because plasma half-life is ~90 min. The highest dose is taken in the morning, the next dose at lunch and the third at afternoon, depending on the patient's needs, in order to replicate cortisol circadian rhythm. Recently a new formulation is available, Plenadren, which may be administered once daily. This formulation consists of two layers, an outer layer responsible for immediate release of the drug, and an internal layer responsible for prolonged release [67]. Open-label studies have shown that it does not affect the BMI of the patients and the lipid profile, but all these findings may be the results of the reduced bioavailability in relation to hydrocortisone [68]. Patients with poor compliance

could be administered prednisolone, which has a longer half-life (150 min) but less mineralocorticoid activity than hydrocortisone. A 40-mg dose of hydrocortisone exerts mineralocorticoid activity equivalent to 100 μg of fludrocortisone. Dexamethasone, because of the complete lack of mineralocorticoid activity, it is not recommended as replacement therapy in patients with adrenal insufficiency. Chronocort, is a newly formulation under development, which tries to mimic the circadian rhythm [69]. For patients with CAH, a CRFR1 antagonist is under study, with the aim to reduce ACTH secretion and minimize glucocorticoid replacement treatment [70].

Plenadren is a modified-release tablet of hydrocortisone that releases hydrocortisone over a longer period of time, allowing once daily dosing. Chronocort is modified-release tablet of hydrocortisone, designed for patients with CAH and in order to mimic the circadian rhythm of cortisol, given twice daily. CRF is the primary regulator of the hypothalamic-pituitary axis, acting directly on specific receptors on pituitary corticotroph cells for ACTH production. CRF antagonists have shown to reduce ACTH release both in vivo and in vitro.

Mineralocorticoid Replacement

Administration of mineralocorticoids is necessary for patients with impaired aldosterone secretion such as Addison disease or the salt-wasting form of CAH or after bilateral adrenalectomy. The therapeutic objective is to maintain normal blood pressure and normal electrolyte levels and to prevent salt craving [71]. Account should be taken of that ~13% of patients with Addison disease show residual aldosterone secretion. The formulation commonly used today is 9α-fluorocortisol (fludrocortisone) which has a 200–400-fold higher mineralocorticoid potency than hydrocortisone [72]. Fludrocortisone exerts its action through specific mineralocorticoid receptors sparse in different tissues, and is very effective to restore electrolyte balance and hypotension. The proper dosage of daily fludrocortisones is achieved by measuring blood pressure, examining for the presence of peripheral oedema and by measuring serum potassium and sodium levels. Renin determination could be useful for dose monitor, but it should take in consideration that it is affected by the use of oestrogens [73]. Signs of mineralocorticoid overtreatment are, high blood pressure, rapid weight gain, oedema and hypokalaemia. Monitoring of the results of the dose changing should be done 2-4 weeks later.

Patients with PAI under treatment with phenytoin, phenobarbital, rifampicin and mitotane, need increased doses of fludrocortisone and hydrocortisone, because the CYP3A4 induction by the drugs, results in rapid inactivation of cortisol (Table 3). In areas with hot climates, especially in the summer months, an increase in mineralocorticoid replacement may be necessary [74].

In the case that the patient under mineralocorticoid develops hypertension, the dosage of glucocorticoids and mineralocorticoids should be reassessed. Antihypertensive treatment of choice in patients with PAI, are direct vasodilator (calcium antagonist), while diuretics and especially aldosterone antagonists (spironolactone or eplerenone), should be avoided. In patients with PAI the occurrence of heart failure can be treated with angiotensin-converting enzyme inhibitors, angiotensin II antagonists or β-blockers, while administration of mineralocorticoid receptor antagonists and/or discontinuation of fludrocortisone should be individualized [75].

DHEA Replacement

DHEA replacement does not seem to be necessary in patients with adrenal insufficiency. Various studies have shown that they can have beneficial effects on mood and on libido (specifically in women) [76]. There is some evidence that oral DHEA possesses a role in immunomodulation, restoring the levels of regulatory T cells in patients with Addison disease. Administration of DHEA is personalised and targeted mainly in women with impaired well being, reduced libido and dry skin.

E. ADRENAL CRISIS

Adrenal crisis may be the first symptom of adrenal insufficiency, or may be the result of inadequate treatment in patients with known adrenal insufficiency when the demand of the organism for glucocorticoids is greater than their availability. Adrenal crisis is more common among patients with primary adrenal insufficiency than in those with secondary adrenal insufficiency [25,77]. The incidence among patients with known adrenal insufficiency is 5–10 events per 100 patient-years [7]. The mortality of adrenal crisis is rare and is 0.5 per 100 patient-years. Increased risk of premature death is observed among patients < 40 years old [78].

Risk factors for the occurrence of adrenal crisis are older age (>65 years) or adolescence and young adulthood, previous crises, and comorbidities [79,80]. Patients on low-dose, short- acting glucocorticoids are in increased risk for adrenal crisis [81], as well as patients treated with drugs that reduce cortisol production (steroidogenesis inhibitors) or increase the metabolic

clearance of cortisol (compounds that induce CYP3A4). Generally, the risk of adrenal crisis is low among patients receiving glucocorticoids treatment [82]. Special attention should be made in patients with thyroid disease. Initiation of thyroxine treatment or the onset of hyperthyroidism in patients with adrenal insufficiency could precipitate an adrenal crisis, because of more rapid inactivation of cortisol [83].

Precipitating factors

In patients with adrenal insufficiency who are receiving replacement treatment, the precipitating factors that may cause adrenal crisis are any kind of severe acute inflammation or sepsis [84]. The most common cause is gastroenteritis, although there is a percentage of patients where no precipitating factor is found (10%) [85]. In this case there is an imbalance between the availability and the demand for cortisol, due to the reduced absorption of orally administered glucocorticoids in these patients.

Other precipitating factors could be a reduction in the dose of administered glucocorticoids or the use of different glucocorticoid preparation or even an intense psychological stress. The pathophysiology of adrenal crisis is multifactorial and is not fully understood. Glucocorticoids affect the immune system. The inflammatory reaction is characterized by the release of cytokines, IL-1, TNF and IL-6. Under normal conditions these cytokines stimulate hypothalamic-pituitary-adrenal axis and increase cortisol secretion [86]. Increased cortisol levels in turn inhibit further cytokine release and action [87,88]. Cortisol thereby prevents a cytokine cascade due to a hyperinflammatory response [89].

Cortisol deficiency observed in adrenal insufficiency leads to increased inflammatory cytokines. In addition, cortisol deficiency is aggravated by a cytokine-induced glucocorticoid receptor resistance. The hypovolemia seen in primary adrenal insufficiency due to aldosterone deficiency, is further aggravated by the reduced action of catecholamines in blood vessels. The hypotension that occurs may be aggravated by vomiting and diarrhea [90].

Lack of cortisol leads to metabolic alterations such as reduced production of fatty acids from adipose tissue, reduced amino-acid liberation from muscles and reduced gluconeogenesis from liver. The result of these changes leads to decreased energy reserves.

Management of adrenal crisis

If an adrenal crisis is suspected, treatment should be promptly initiated, without waiting for laboratory test results that document cortisol deficiency. A rapid patient's medical history, such as the abrupt discontinuation of chronic exogenous glucocorticoids use, or the occurrence of an infection in patients with known AI, increases the index of suspicion for an adrenal crisis.

Initially must be administered intravenous or intramuscular bolus injection of 100 mg hydrocortisone, followed by continuous intravenous infusion of 200 mg of hydrocortisone per 24 hours, or alternatively, by intravenous or intramuscular bolus injections of 50 mg of hydrocortisone every 6 hours.

Intravenous administration of fluids (isotonic saline), under tight control to avoid either exacerbation or too-rapid correction of hyponatraemia. Sometimes, especially in children it is necessary to treat hypoglycemia. It is very important to identify the precipitating factor and to cure it. Once the recovery of the patient is achieved the hydrocortisone dose should be tapered according to the clinical situation of the patient. A delay of the treatment of an adrenal crisis increases the risk of death. Adrenal insufficiency is a rare disease, but misdiagnosis could be potentially fatal. All patients with AI need education in order to recognize signs and symptoms of adrenal crisis, they must be equipped with a hydrocortisone self-injection kit for emergency management and with a steroid emergency card [91].

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REFERENCES

1. Arlt W, Allolio B. Adrenal insufficiency. Lancet Lond Engl. 2003;361(9372):1881–93.
2. Ross IL, Levitt NS. Addison's disease symptoms--a cross sectional study in urban South Africa. PloS One. 2013;8(1):e53526.
3. Erichsen MM, Løvås K, Skinningsrud B, Wolff AB, Undlien DE, Svartberg J, et al. Clinical, immunological, and genetic features of autoimmune primary adrenal insufficiency: observations from a Norwegian registry. J Clin Endocrinol Metab. 2009;94(12):4882–4890.
4. Odeniyi IA, Fasanmade OA, Ajala MO, Ohwovoriole AE. Adrenocortical function in Nigerians with human immunodeficiency virus infection. Ghana Med J. 2013;47(4):171–7.
5. Broersen LHA, Pereira AM, Jørgensen JOL, Dekkers OM. Adrenal Insufficiency in Corticosteroids Use: Systematic Review and Meta-Analysis. J Clin Endocrinol Metab. 2015;100(6):2171–80.
6. Bleicken B, Hahner S, Ventz M, Quinkler M. Delayed diagnosis of adrenal insufficiency is common: a cross-sectional study in 216 patients. Am J Med Sci. 2010;339(6):525–31.

7. Smans LCCJ, Van der Valk ES, Hermus ARMM, Zelissen PMJ. Incidence of adrenal crisis in patients with adrenal insufficiency. Clin Endocrinol (Oxf). 2016;84(1):17–22.
8. Gjerstad JK, Lightman SL, Spiga F. Role of glucocorticoid negative feedback in the regulation of HPA axis pulsatility. Stress Amst Neth. 2018;21(5):403–16.
9. Roelfsema F, Aoun P, Veldhuis JD. Pulsatile Cortisol Feedback on ACTH Secretion Is Mediated by the Glucocorticoid Receptor and Modulated by Gender. J Clin Endocrinol Metab. 2016;101(11):4094–102.
10. Lightman SL, Conway-Campbell BL. The crucial role of pulsatile activity of the HPA axis for continuous dynamic equilibration. Nat Rev Neurosci. 2010;11(10):710–8.
11. Walker JJ, Terry JR, Lightman SL. Origin of ultradian pulsatility in the hypothalamic-pituitary-adrenal axis. Proc Biol Sci. 2010;277(1688):1627–33.
12. Oster H, Challet E, Ott V, Arvat E, de Kloet ER, Dijk DJ, et al. The Functional and Clinical Significance of the 24-Hour Rhythm of Circulating Glucocorticoids. Endocr Rev. 2017;38(1):3–45.
13. Caratti G, Matthews L, Poolman T, Kershaw S, Baxter M, Ray D. Glucocorticoid receptor function in health and disease. Clin Endocrinol (Oxf). 2015;83(4):441–8.
14. Chan WL, Carrell RW, Zhou A, Read RJ. How changes in affinity of corticosteroid-binding globulin modulate free cortisol concentration. J Clin Endocrinol Metab. 2013;98(8):3315–22.
15. Henley D, Lightman S, Carrell R. Cortisol and CBG - Getting cortisol to the right place at the right time. Pharmacol Ther. 2016;166:128–35.
16. Tucci JR, Espiner EA, Jagger PI, Pauk GL, Lauler DP. ACTH stimulation of aldosterone secretion in normal subjects and in patients with chronic adrenocortical insufficiency. J Clin Endocrinol Metab. 1967;27(4):568–75.
17. Kem DC, Weinberger MH, Higgins JR, Kramer NJ, Gomez-Sanchez C, Holland OB. Plasma aldosterone response to ACTH in primary aldosteronism and in patients with low renin hypertension. J Clin Endocrinol Metab. 1978;46(4):552–60.
18. Bollag WB. Regulation of aldosterone synthesis and secretion. Compr Physiol. 2014;4(3):1017–55.
19. Afreen B, Khan KA, Riaz A. Adrenal Insufficiency In Pakistani Hiv Infected Patients. J Ayub Med Coll Abbottabad JAMC. 2017;29(3):428–31.
20. Mofokeng TRP, Beshyah SA, Mahomed F, Ndlovu KCZ, Ross IL. Significant barriers to diagnosis and management of adrenal insufficiency in Africa. Endocr Connect. 2020;9(5):445–56.
21. Tripathy SK, Agrawala RK, Baliarsinha AK. Endocrine alterations in HIV-infected patients. Indian J Endocrinol Metab. 2015;19(1):143–7.
22. Hellesen A, Bratland E, Husebye ES. Autoimmune Addison's disease - An update on pathogenesis. Ann Endocrinol. 2018;79(3):157–63.
23. Laureti S, Vecchi L, Santeusanio F, Falorni A. Is the prevalence of Addison's disease underestimated? J Clin Endocrinol Metab. 1999;84(5):1762.
24. Björnsdottir S, Sundström A, Ludvigsson JF, Blomqvist P, Kämpe O, Bensing S. Drug prescription patterns in patients with Addison's disease: a Swedish population-based cohort study. J Clin Endocrinol Metab. 2013;98(5):2009–18.
25. Meyer G, Neumann K, Badenhoop K, Linder R. Increasing prevalence of Addison's disease in German females: health insurance data 2008-2012. Eur J Endocrinol. 2014;170(3):367–73.
26. Stewart PM, Biller BMK, Marelli C, Gunnarsson C, Ryan MP, Johannsson G. Exploring Inpatient Hospitalizations and Morbidity in Patients With Adrenal Insufficiency. J Clin Endocrinol Metab. 2016;101(12):4843–50.
27. Chantzichristos D, Persson A, Eliasson B, Miftaraj M, Franzén S, Svensson AM, et al. Incidence, prevalence and seasonal onset variation of Addison's disease among persons with type 1 diabetes mellitus: nationwide, matched cohort studies. Eur J Endocrinol. 2018;178(1):113–20.
28. Bancos I, Hahner S, Tomlinson J, Arlt W. Diagnosis and management of adrenal insufficiency. Lancet Diabetes Endocrinol. 2015;3(3):216–26.
29. White PC, Speiser PW. Congenital adrenal hyperplasia due to 21-hydroxylase deficiency. Endocr Rev. 2000;21(3):245–91.
30. Merke DP, Auchus RJ. Congenital Adrenal Hyperplasia Due to 21-Hydroxylase Deficiency. N Engl J Med. 2020;383(13):1248–61.
31. Dalin F, Nordling Eriksson G, Dahlqvist P, Hallgren Å, Wahlberg J, Ekwall O, et al. Clinical and Immunological Characteristics of Autoimmune Addison Disease: A Nationwide Swedish Multicenter Study. J Clin Endocrinol Metab. 2017;102(2):379–89.
32. Mitchell AL, Pearce SHS. Autoimmune Addison disease: pathophysiology and genetic complexity. Nat Rev Endocrinol. 2012;8(5):306–16.
33. Bratland E, Skinningsrud B, Undlien DE, Mozes E, Husebye ES. T cell responses to steroid cytochrome P450 21-hydroxylase in patients with autoimmune primary adrenal insufficiency. J Clin Endocrinol Metab. 2009;94(12):5117–24.
34. Naletto L, Frigo AC, Ceccato F, Sabbadin C, Scarpa R, Presotto F, et al. The natural history of autoimmune Addison's disease from the detection of autoantibodies to development of the disease: a long-term follow-up study on 143 patients. Eur J Endocrinol. 2019;180(3):223–34.
35. Eriksson D, Bianchi M, Landegren N, Dalin F, Skov J, Hultin-Rosenberg L, et al. Common genetic variation in the autoimmune regulator (AIRE) locus is associated with autoimmune Addison's disease in Sweden. Sci Rep. 2018;8(1):8395.
36. Skov J, Höijer J, Magnusson PKE, Ludvigsson JF, Kämpe O, Bensing S. Heritability of Addison's disease and prevalence of associated autoimmunity in a cohort of 112,100 Swedish twins. Endocrine. 2017;58(3):521–7.
37. Loeb RF. Chemical changes in the blood in Addison's disease. Science. 1932;76(1975):420–1.
38. Regal M, Páramo C, Sierra SM, Garcia-Mayor RV. Prevalence and incidence of hypopituitarism in an adult Caucasian population in northwestern Spain. Clin Endocrinol (Oxf). 2001;55(6):735–40.
39. Bates AS, Van't Hoff W, Jones PJ, Clayton RN. The effect of hypopituitarism on life expectancy. J Clin Endocrinol Metab. 1996;81(3):1169–72.

40. Jenkins PJ, Bates P, Carson MN, Stewart PM, Wass JAH. Conventional pituitary irradiation is effective in lowering serum growth hormone and insulin-like growth factor-I in patients with acromegaly. J Clin Endocrinol Metab. 2006;91(4):1239–45.
41. Appelman-Dijkstra NM, Kokshoorn NE, Dekkers OM, Neelis KJ, Biermasz NR, Romijn JA, et al. Pituitary dysfunction in adult patients after cranial radiotherapy: systematic review and meta-analysis. J Clin Endocrinol Metab. 2011;96(8):2330–40.
42. Knappe UJ, Petroff D, Quinkler M, Schmid SM, Schöfl C, Schopohl J, et al. Fractionated radiotherapy and radiosurgery in acromegaly: analysis of 352 patients from the German Acromegaly Registry. Eur J Endocrinol. 2020;182(3):275–84.
43. Turcu AF, Erickson BJ, Lin E, Guadalix S, Schwartz K, Scheithauer BW, et al. Pituitary stalk lesions: the Mayo Clinic experience. J Clin Endocrinol Metab. 2013;98(5):1812–8.
44. Barroso-Sousa R, Barry WT, Garrido-Castro AC, Hodi FS, Min L, Krop IE, et al. Incidence of Endocrine Dysfunction Following the Use of Different Immune Checkpoint Inhibitor Regimens: A Systematic Review and Meta-analysis. JAMA Oncol. 2018;4(2):173–82.
45. de Vries F, Bruin M, Lobatto DJ, Dekkers OM, Schoones JW, van Furth WR, et al. Opioids and Their Endocrine Effects: A Systematic Review and Meta-analysis. J Clin Endocrinol Metab. 2020;105(3):1020–9.
46. Fountas A, Van Uum S, Karavitaki N. Opioid-induced endocrinopathies. Lancet Diabetes Endocrinol. 2020;8(1):68–80.
47. Woods CP, Argese N, Chapman M, Boot C, Webster R, Dabhi V, et al. Adrenal suppression in patients taking inhaled glucocorticoids is highly prevalent and management can be guided by morning cortisol. Eur J Endocrinol. 2015;173(5):633–42.
48. Mebrahtu TF, Morgan AW, Keeley A, Baxter PD, Stewart PM, Pujades-Rodriguez M. Dose dependency of iatrogenic glucocorticoid excess and adrenal insufficiency and mortality: a cohort study in England. J Clin Endocrinol Metab. 2019;104(9):3757–67.
49. Joseph RM, Hunter AL, Ray DW, Dixon WG. Systemic glucocorticoid therapy and adrenal insufficiency in adults: A systematic review. Semin Arthritis Rheum. 2016;46(1):133–41.
50. Leong SH, Shander S, Ratnasingam J. Predicting recovery of the hypothalamic-pituitary-adrenal axis after prolonged glucocorticoid use. Endocr Pract Off J Am Coll Endocrinol Am Assoc Clin Endocrinol. 2018;24(1):14–20.
51. Tutunculer F, Saka N, Arkaya SC, Abbasoglu S, Bas F. Evaluation of adrenomedullary function in patients with congenital adrenal hyperplasia. Horm Res. 2009;72(6):331–6.
52. Geiger AM, Pitts KP, Feldkamp J, Kirschbaum C, Wolf JM. Cortisol-dependent stress effects on cell distribution in healthy individuals and individuals suffering from chronic adrenal insufficiency. Brain Behav Immun. 2015;50:241–248.
53. Ehrhart-Bornstein M, Hinson JP, Bornstein SR, Scherbaum WA, Vinson GP. Intraadrenal interactions in the regulation of adrenocortical steroidogenesis. Endocr Rev. 1998;19(2):101–43.
54. Weise M, Mehlinger SL, Drinkard B, Rawson E, Charmandari E, Hiroi M, et al. Patients with classic congenital adrenal hyperplasia have decreased epinephrine reserve and defective glucose elevation in response to high-intensity exercise. J Clin Endocrinol Metab. 2004;89(2):591–7.
55. Elbelt U, Hahner S, Allolio B. Altered insulin requirement in patients with type 1 diabetes and primary adrenal insufficiency receiving standard glucocorticoid replacement therapy. Eur J Endocrinol. 2009;160(6):919–24.
56. Labrie F, Simard J, Luu-The V, Pelletier G, Belghmi K, Bélanger A. Structure, regulation and role of 3 beta-hydroxysteroid dehydrogenase, 17 beta-hydroxysteroid dehydrogenase and aromatase enzymes in the formation of sex steroids in classical and peripheral intracrine tissues. Baillieres Clin Endocrinol Metab. 1994;8(2):451–74.
57. El-Farhan N, Pickett A, Ducroq D, Bailey C, Mitchem K, Morgan N, et al. Method-specific serum cortisol responses to the adrenocorticotrophin test: comparison of gas chromatography-mass spectrometry and five automated immunoassays. Clin Endocrinol (Oxf). 2013;78(5):673–80.
58. Ueland GÅ, Methlie P, Øksnes M, Thordarson HB, Sagen J, Kellmann R, et al. The Short Cosyntropin Test Revisited: New Normal Reference Range Using LC-MS/MS. J Clin Endocrinol Metab. 2018;103(4):1696–703.
59. Dickstein G, Shechner C, Nicholson WE, Rosner I, Shen-Orr Z, Adawi F, et al. Adrenocorticotropin stimulation test: effects of basal cortisol level, time of day, and suggested new sensitive low dose test. J Clin Endocrinol Metab. 1991;72(4):773–8.
60. Mao JJ, Dages KN, Suresh M, Bancos I. Presentation, disease progression and outcomes of adrenal gland metastases. Clin Endocrinol (Oxf). 2020;93(5):546–54.
61. Grossman AB. Clinical Review#: The diagnosis and management of central hypoadrenalism. J Clin Endocrinol Metab. 2010;95(11):4855–63.
62. Erturk E, Jaffe CA, Barkan AL. Evaluation of the integrity of the hypothalamic-pituitary-adrenal axis by insulin hypoglycemia test. J Clin Endocrinol Metab. 1998;83(7):2350–4.
63. Mukherjee JJ, de Castro JJ, Kaltsas G, Afshar F, Grossman AB, Wass JA, et al. A comparison of the insulin tolerance/glucagon test with the short ACTH stimulation test in the assessment of the hypothalamo-pituitary-adrenal axis in the early post-operative period after hypophysectomy. Clin Endocrinol (Oxf). 1997;47(1):51–60.
64. Steiner H, Bähr V, Exner P, Oelkers PW. Pituitary function tests: comparison of ACTH and 11-deoxy-cortisol responses in the metyrapone test and with the insulin hypoglycemia test. Exp Clin Endocrinol. 1994;102(1):33–8.
65. Debono M, Harrison RF, Whitaker MJ, Eckland D, Arlt W, Keevil BG, et al. Salivary Cortisone Reflects Cortisol Exposure Under Physiological Conditions and After Hydrocortisone. J Clin Endocrinol Metab. 2016;101(4):1469–77.
66. Bornstein SR, Allolio B, Arlt W, Barthel A, Don-Wauchope A, Hammer GD, et al. Diagnosis and Treatment of Primary Adrenal Insufficiency: An Endocrine Society Clinical Practice Guideline. J Clin Endocrinol Metab. 2016;101(2):364–89.
67. Johannsson G, Nilsson AG, Bergthorsdottir R, Burman P,

Dahlqvist P, Ekman B, et al. Improved cortisol exposure-time profile and outcome in patients with adrenal insufficiency: a prospective randomized trial of a novel hydrocortisone dual-release formulation. J Clin Endocrinol Metab. 2012;97(2):473–81.
68. Giordano R, Guaraldi F, Marinazzo E, Fumarola F, Rampino A, Berardelli R, et al. Improvement of anthropometric and metabolic parameters, and quality of life following treatment with dual-release hydrocortisone in patients with Addison's disease. Endocrine. 2016;51(2):360–8.
69. Whitaker M, Debono M, Huatan H, Merke D, Arlt W, Ross RJ. An oral multiparticulate, modified-release, hydrocortisone replacement therapy that provides physiological cortisol exposure. Clin Endocrinol (Oxf). 2014;80(4):554–61.
70. Turcu AF, Spencer-Segal JL, Farber RH, Luo R, Grigoriadis DE, Ramm CA, et al. Single-Dose Study of a Corticotropin-Releasing Factor Receptor-1 Antagonist in Women With 21-Hydroxylase Deficiency. J Clin Endocrinol Metab. 2016;101(3):1174–80.
71. Pofi R, Prete A, Thornton-Jones V, Bryce J, Ali SR, Faisal Ahmed S, et al. Plasma Renin Measurements are Unrelated to Mineralocorticoid Replacement Dose in Patients With Primary Adrenal Insufficiency. J Clin Endocrinol Metab. 2020;105(1):dgz055.
72. Oelkers W, Diederich S, Bähr V. Diagnosis and therapy surveillance in Addison's disease: rapid adrenocorticotropin (ACTH) test and measurement of plasma ACTH, renin activity, and aldosterone. J Clin Endocrinol Metab. 1992;75(1):259–64.
73. Flad TM, Conway JD, Cunningham SK, McKenna TJ. The role of plasma renin activity in evaluating the adequacy of mineralocorticoid replacement in primary adrenal insufficiency. Clin Endocrinol (Oxf). 1996;45(5):529–34.
74. Chortis V, Taylor AE, Schneider P, Tomlinson JW, Hughes BA, O'Neil DM, et al. Mitotane therapy in adrenocortical cancer induces CYP3A4 and inhibits 5α-reductase, explaining the need for personalized glucocorticoid and androgen replacement. J Clin Endocrinol Metab. 2013;98(1):161–71.
75. Esposito K, Maiorino MI, Bellastella G, Giugliano F, Romano M, Giugliano D. Determinants of female sexual dysfunction in type 2 diabetes. Int J Impot Res. 2010;22(3):179–84.
76. Gurnell EM, Hunt PJ, Curran SE, Conway CL, Pullenayegum EM, Huppert FA, et al. Long-term DHEA replacement in primary adrenal insufficiency: a randomized, controlled trial. J Clin Endocrinol Metab. 2008;93(2):400–9.
77. Ritzel K, Beuschlein F, Mickisch A, Osswald A, Schneider HJ, Schopohl J, et al. Clinical review: Outcome of bilateral adrenalectomy in Cushing's syndrome: a systematic review. J Clin Endocrinol Metab. 2013;98(10):3939–48.
78. Erichsen MM, Løvås K, Fougner KJ, Svartberg J, Hauge ER, Bollerslev J, et al. Normal overall mortality rate in Addison's disease, but young patients are at risk of premature death. Eur J Endocrinol. 2009;160(2):233–7.
79. Hahner S, Loeffler M, Bleicken B, Drechsler C, Milovanovic D, Fassnacht M, et al. Epidemiology of adrenal crisis in chronic adrenal insufficiency: the need for new prevention strategies. Eur J Endocrinol. 2010;162(3):597–602.
80. El-Maouche D, Hargreaves CJ, Sinaii N, Mallappa A, Veeraraghavan P, Merke DP. Longitudinal Assessment of Illnesses, Stress Dosing, and Illness Sequelae in Patients With Congenital Adrenal Hyperplasia. J Clin Endocrinol Metab. 2018;103(6):2336–45.
81. Rushworth RL, Torpy DJ. Adrenal Insufficiency in Australia: Is it Possible that the Use of Lower Dose, Short-Acting Glucocorticoids has Increased the Risk of Adrenal Crises? Horm Metab Res Horm Stoffwechselforschung Horm Metab. 2015;47(6):427–32.
82. Rushworth RL, Chrisp GL, Torpy DJ. Glucocorticoid-induced adrenal insufficiency: A study of the incidence in hospital patients and a review of peri-operative management. Endocr Pract Off J Am Coll Endocrinol Am Assoc Clin Endocrinol. 2018;24(5):437–5.
83. Fonseca V, Brown R, Hochhauser D, Ginsburg J, Havard CW. Acute adrenal crisis precipitated by thyroxine. Br Med J Clin Res Ed. 1986;292(6529):1185–6.
84. Puar THK, Stikkelbroeck NMML, Smans LCCJ, Zelissen PMJ, Hermus ARMM. Adrenal Crisis: Still a Deadly Event in the 21st Century. Am J Med. 2016;129(3):339.e1-9.
85. Hahner S, Spinnler C, Fassnacht M, Burger-Stritt S, Lang K, Milovanovic D, et al. High incidence of adrenal crisis in educated patients with chronic adrenal insufficiency: a prospective study. J Clin Endocrinol Metab. 2015;100(2):407–16.
86. Besedovsky H, del Rey A, Sorkin E, Dinarello CA. Immunoregulatory feedback between interleukin-1 and glucocorticoid hormones. Science. 1986;233(4764):652–4.
87. Barber AE, Coyle SM, Marano MA, Fischer E, Calvano SE, Fong Y, et al. Glucocorticoid therapy alters hormonal and cytokine responses to endotoxin in man. J Immunol Baltim Md 1950. 1993;150(5):1999–2006.
88. Morrow LE, McClellan JL, Conn CA, Kluger MJ. Glucocorticoids alter fever and IL-6 responses to psychological stress and to lipopolysaccharide. Am J Physiol. 1993;264(5 Pt 2):R1010-6.
89. Sapolsky RM, Romero LM, Munck AU. How do glucocorticoids influence stress responses? Integrating permissive, suppressive, stimulatory, and preparative actions. Endocr Rev. 2000;21(1):55–89.
90. Quinkler M, Oelkers W, Remde H, Allolio B. Mineralocorticoid substitution and monitoring in primary adrenal insufficiency. Best Pract Res Clin Endocrinol Metab. 2015;29(1):17–24.
91. Allolio B. Extensive expertise in endocrinology. Adrenal crisis. Eur J Endocrinol. 2015;172(3):R115-124.

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COVER LETTER

A submission letter to the Editor should accompany the manuscript and contain the following:

- The manuscript has not been published previously, and is not under consideration for publication elsewhere.
- Acknowledgment of grants or financial support.
- The manuscript has been approved by all authors.

INFORMATION ABOUT ARTICLE TYPES

The Editors will consider and publish the following:

1. Original research articles
2. Narrative Reviews
3. Systematic Reviews and Meta-analyses
4. Editorials
5. Letters to the Editor
6. Case Reports

Original research articles

The maximum length of the main text is 3,500 words excluding the abstract, references, tables, and figure legends. A maximum of 6 tables and/or figures is allowed. References should not exceed a maximum of 100.

Narrative Reviews / Systematic Reviews / Meta-analyses

These manuscripts are solicited and unsolicited manuscripts that feature an organized and detailed review of the scientific literature about a particular topic. This section is peer-reviewed and acceptance for publication is not guaranteed. The maximum length of the main text is 5,000 words excluding the abstract, references, tables, and figure legends. A maximum of 6 tables and/or figures to summarize critical points is highly desirable. References should not exceed a maximum of 150.

Editorials

Editorials are usually solicited by the Editor. The maximum length is 1500 words excluding the references, tables, and figure legends. One table or 1 figure is allowed. References should not exceed a maximum of 20. Editorials may have a maximum of three (3) authors.

Letters to the Editor

Letters to the Editor will be considered for publication if they are related to articles published in recent issues of the Achaiki Iatriki Journal. The maximum length is 800 words (excluding references, table, and figure legend). A total number of 1 table or figure is allowed and up to 10 references. Such letters will be passed to the authors of the original paper, who will be offered an opportunity to reply. Letters to the Editor may have a maximum of two (2) authors.

Case Reports

Case reports should ideally include a short introduction, the case presentation and a brief discussion. The maximum length is 1500 words (excluding references, tables, and figure legend). A total number of 2 tables or figures is allowed. References should not exceed a maximum of 15.

Formatting guide

The articles must by typewritten and double spaced. They should include the following sections, each starting on a separate page:

- Title Page
- Abstract and Key Words
- Main Text
- Acknowledgements
- References
- Tables
- Figures

Margins should be not less than 2.5 cm. Pages should be numbered consecutively.

Abbreviations

Do not use non-standard abbreviations. The use of abbreviations in the title and abstract should be avoided. Abbreviations should be defined on their first appearance in the text; those not accepted by international bodies should be avoided.

Title page

The title page should include:

- Title of the manuscript
- Short title which will be used as a running head
- Full name of each author
- Full location of department and institution where work was performed
- Name and address for correspondence, including fax number, telephone number, and e-mail address.
- Conflict of interest disclosure.
- Declaration of funding sources.
- Author Contributions according to the following criteria for authorship: conception and design; analysis and interpretation of the data; drafting of the article; critical revision of the article for important intellectual content; final approval of the article.

Abstract

For Original Articles, structured abstracts should be 250 words or less and include the following sections: Background, Methods, Results and Conclusion. Review articles should carry an unstructured abstract which should not exceed 200 words.

Key words

The abstract should be followed by a list of 3–5 keywords which will assist the cross-indexing of the article and which may be published separated by semicolons.

Main Text

For the main body of the text, the recommended structure of

the manuscript is:
- Introduction
- Materials and Methods
- Results
- Discussion

Define abbreviations at first mention in text and in each table and figure.

Introduction

State the objectives of the work and provide an adequate background, avoiding a detailed literature survey or a summary of the results.

Materials and Methods

Provide sufficient detail to allow the work to be reproduced. Methods already published should be indicated by a reference. This includes a description of the design, measurement and collection of data, type and source of subjects, inclusion and exclusion criteria and measures of outcome, number of subjects studied and why this number was chosen. Any deviation from the study protocol should be stated. Randomized controlled trials should adhere to the CONSORT guidelines that can be found at: http://www.consort-statement.org. Observational studies should also adhere to Strobe statement: http://www.strobe-statement.org/. Diagnostic accuracy studies should follow the Stard statement: http://www.stard-statement.org/. Systematic Reviews and Meta-Analyses should adhere to the PRISMA statement: http://www.prisma-statement.org/.

Statistical analysis

The statistical methods used should be relevant and clearly stated. Special or complex statistical methods should be explained and referenced. Describe statistical methods with enough detail to enable a knowledgeable reader with access to the original data to verify the reported results. When possible, quantify findings and present them with appropriate indicators of measurement error or uncertainty (such as confidence intervals). Avoid relying solely on statistical hypothesis testing, such as P values, which fail to convey important information about effect size. Define statistical terms, abbreviations, and symbols. Specify the software used.

Units

Follow internationally accepted rules and conventions: use the internal system of units (SI).

Results

Results should be clear and concise. Results should be explained and illustrated by using Tables and Figures. Do not duplicate information contained in tables and figures.

Discussion

Discussion should directly relate to the results of the study and should explore their significance. Do not provide a general review of the topic.

Conclusions

The conclusions should provide a summary of the key results and discuss the appropriateness and impact of this original work.

Acknowledgements

Collate acknowledgements in a separate section at the end of the article before the references. Acknowledgements should be made only to those who have made a substantial contribution to the study. Authors are responsible for obtaining written permission from people acknowledged by name in case readers infer their endorsement of data and conclusions.

References

Ensure that every reference cited in the text is also present in the reference list (and vice versa). References should be numbered in the order they appear in the text. Manuscripts should follow the style of the Vancouver agreement detailed in the International Committee of Medical Journal Editors' revised 'Uniform Requirements for Manuscripts Submitted to Biomedical Journals: Writing and Editing for Biomedical Publication', as presented at http://www.ICMJE.org/. In the text, references should be cited using Arabic numerals enclosed in square brackets [1]. The last names and initials of all authors should be referred to if they are up to six, otherwise only the first six are referred, with et al following. References should also include full title and source information. Journal names should be abbreviated according to the standard in the Index Medicus. No periods should be placed at the end of abbreviations of the journal.

Journal article, up to 6 personal author(s):

Example: Al-Habian A, Harikumar PE, Stocker CJ, Langlands K, Selway JL. Histochemical and immunohistochemical evaluation of mouse skin histology: comparison of fixation with neutral buffered formalin and alcoholic formalin. J Histotechnol. 2014;37(4):115-24.

Journal article, more than 6 personal author(s):

Example: Liaw S, Hasan I, Wade, V, Canalese R, Kelaher M, Lau P, et al. Improving cultural respect to improve Aboriginal health in general practice: a multi-perspective pragmatic study. Aust Fam Physician. 2015;44(6):387-92.

Journal article/ Issue with a supplement

Example: Bonda C, Sharma P, LaFaver K. Clinical reasoning: a 28 year-old woman with lower extremity spasticity and microcytic anemia. Neurology. 2015;85(2) Suppl:e11-4.

Electronic journal article:

Example: Poling J, Kelly L, Chan C, Fisman D, Ulanova M. Hospital admission for community-acquired pneumonia in a First Nations population. Can J Rural Med [Internet]. 2014 Fall [cited 2015 Apr 27];19(4):135-41. Available from: http://www.srpc.ca/14fal.html by selecting PDF link in table of contents.

Book, personal author(s):

Example: Buckingham L. Molecular diagnostics: fundamentals, methods and clinical applications. 2nd ed. Philadelphia: F.A. Davis; c2012.

Book or pamphlet, organization as both author and publisher:

Example: College of Medical Radiation Technologists of Ontario. Standards of practice. Toronto: The College; 2011.

Book, editor(s):

Example: Kumar V, Abbas AK, Aster JC, editors. Robbins basic pathology. 16th ed. Philadelphia: Elsevier Saunders; c2013.

Poster presentation/session presented at a meeting or conference:

Example: Chasman J, Kaplan RF. The effects of occupation on preserved cognitive functioning in dementia. Poster session presented at: Excellence in clinical practice. 4th Annual Conference of the American Academy of Clinical Neuropsychology; 2006 Jun 15-17; Philadelphia, PA.

Tables

Tables should be typewritten, double-spaced, each one on a separate page and numbered consecutively with Arabic numerals in the order of their appearance in the text. Do not duplicate material presented in a figure. Tables should include a short but concise title. Tables should read vertically when possible. Place explanatory matter in footnotes, including any non-standard abbreviation. If data from another published or unpublished source are used, obtain permission and acknowledge fully.

Figure legends

Figure legends should be listed one after the other, as part of the main text, separate from the figure files. Each figure legend should have a brief title (in bold with figure number) followed by a description of each panel, and the symbols used. The statistical test used as well as the values of statistical significance (whether significant or not) should always be included in the figure legends. If a figure has been published previously, acknowledge the original source and submit written permission from the copyright holder to reproduce it. Authors will be required to pay for the extra cost of printing illustrations in color. However, there is an option to have their images in color in the electronic version of their manuscript and in grey scale in the printed version.

Figures

All figures for review should be submitted as a separate file in JPEG or TIFF format in grayscales or in RGB color mode with a resolution of at least 300 dpi. Number figures consecutively using Arabic numerals.

Photographs should be submitted as TIFF with a resolution of at least 300 pixels per inch; or Illustrator compatible EPS files with RGB color management or Photoshop or editable PDF files (grayscales or RGB).

Photographs of identifiable patients should be accompanied by written permission to publish from patient(s).

RGB figures will be presented in color in the electronic version and in grey scale in the printed version.

Ethical Considerations

An author should not publish manuscripts describing essentially the same research in more than one journal or primary publication. It must not be under consideration for publication elsewhere, and, if accepted, must not be published elsewhere in similar form, in any language. The International Committee of Medical Journal Editors has a full description about duplicate or redundant publication (http://www.icmje.org).

Authorship should be limited to those who have made a significant contribution to the conception, design, execution, or interpretation of the reported study.

The 'Achaiki Iatriki' editors endorse the principles of the Declaration of Helsinki and expect that all investigations involving humans will have been performed in accordance with these principles.

Authors should carefully protect patients' anonymity. Manuscripts reporting data from research conducted on humans must include a statement of assurance in the materials and methods section describing that: written informed consent was obtained from each patient included in the study and that the study protocol conforms to the ethical guidelines of the 1975 Declaration of Helsinki as reflected in a priori approval by the institution's human research committee.

Do not use patients' names, initials, or hospital numbers, especially in illustrative material.

For animal experimentation reported in the journal, it is expected that investigators will have observed the Interdisciplinary Principles and Guidelines for the Use of Animals in Research, Testing, and Education issued by the New York Academy of Sciences' Adhoc Committee on Animal Research.

Disclosures: Conflict of interest

All authors are required to provide a Declaration of Interest Statement recognizing and disclosing financial and other conflicts of interest that might bias their work. Particularly, they disclose any actual or potential conflict of interest including any financial, activities, additional affiliations, personal or other relationships with other people or organizations within three years of beginning the submitted work that could inappropriately influence, or be perceived to influence, their work. Further information at International Committee of Medical Journal Editors ("Uniform Requirements for Manuscripts Submitted to Biomedical Journals") -- February 2006

Disclosures: Financial disclosure

Authors are requested to identify who provided financial support for the conduct of the research and/or preparation of the article and to briefly describe the role of the sponsor(s), if any, in study design; in the collection, analysis and interpretation of data; in the writing of the report; and in the decision to submit the article for publication. If the funding source(s) had no such involvement then this should be stated.

Inform Consent

Patients have a right to privacy that should not be infringed without informed consent. Information such as patients' names, initials, or hospital numbers, should not be published in written descriptions, photographs, and pedigrees unless the information is essential for scientific purposes and the patient (or parent or guardian) gives written informed consent.

Identifying details should be omitted if they are not essential. Complete anonymity is difficult to achieve, however, and informed consent should be obtained if there is any doubt. For example, masking the eye region in photographs of patients is inadequate protection of anonymity. If identifying characteristics are altered to protect anonymity, such as in genetic pedigrees, authors should provide assurance that alterations do not distort scientific meaning.

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Journal Editors ("Uniform Requirements for Manuscripts Submitted to Biomedical Journals") -- February 2006

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Manuscripts reporting experiments using humans or animals must include a statement giving assurance that all humans or animals received human care and that study protocols comply with the institution's guidelines. When reporting experiments on human subjects, authors should indicate whether the procedures followed were in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1975, as revised in 2000. When reporting experiments on animals, authors should be asked to indicate whether the institutional and national guide for the care and use of laboratory animals was followed.

Further information at International Committee of Medical Journal Editors ("Uniform Requirements for Manuscripts Submitted to Biomedical Journals") -- February 2006

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MANUSCRIPT PROCESSING AND REVIEW

Submission

You can submit your manuscript either in Journal's website submission system or via emaill to achaiki.iatriki@gmail.com

Review process

Each manuscript submitted to ACHAIKI IATRIKI is assigned to a Section Editor who has expertise on the subject of the manuscript. The Section Editor initially evaluates the manuscript if it is appropriate and competitive for publication and sends the manuscript to 2-4 reviewers who are experts in the field.

PUBLICATION

Proofs

Proofs will be made available to the author(s) to be checked. It is the responsibility of the author(s) to make sure that the quality and accuracy of the manuscript, figures, and tables in the proofs is correct. At this stage, authors may make only minor corrections. Authors should return their proofs within 48 hours, by e-mail. At this point the author may order reprints, which are charged according to the number of reprints and the number of pages of the article.

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