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Non Celiac Gluten Sensitivity - A Literature Review

Dr Parvathi S Hari[1], Dr Gayathri S Hari[2], Dr Sudha P[3]
Intern, Government Medical College, Thiruvananthapuram, Kerala, India.
2017 Mbbs Batch, Government Medical College, Thiruvananthapuram, Kerala, India.
Additional Professor, Regional Cancer Centre, Thiruvananthapuram, Kerala, India.

Abstract
Non celiac gluten Sensitivity (NCGS) is a largely ambiguous group of disorders with evolving epidemiology. According to the Oslo definition, NCGS consists of "a variety of immunological, morphological, or symptomatic manifestations that are precipitated by the ingestion of gluten in individuals in whom celiac disease has been excluded" [5]. It is characterised by both intestinal and extra intestinal symptoms. The pathophysiology of NCGS is still unclear. Some studies point towards a multifactorial etiopathogenesis of this clinical condition which included immune response induced on exposure to gluten, microbiota shifts and functional effect of fermentable oligosaccharides, disaccharides, monosaccharides and polyols (FODMAP)[15].The levels of CD14 lymphocytes and lipopolysaccharide binding protein[7] was found to be higher in NCGS when compared to healthy controls and Celiac disease(CD). Studies also speculate that the expression of interferon gamma transglutinase 2 in the intestinal mucosa in NCGS plays a role in its pathogenesis. It has been postulated that gliadin may induce mast cell degranulation which leads to inflammatory cytokine production resulting in a low grade intestinal inflammation. Some studies speculate the role of gut microbiota in the pathogenesis of the condition. Kabbani et al devised an algorithm for the diagnosis of this new clinical entity which resulted in identifying CD in 7% of individuals with suspected NCGS while confirming NCGS in the remaining 93%. "Salerno criteria" [6] is now being used to arrive at a diagnosis. There are no available biomarkers or other reliable diagnostic tests that will help to detect this disorder. Therefore the diagnosis is based on clinical exclusion. Gluten free diet, is being used since 1941 for the treatment of gluten related disorders. Long term strict adherence to GFD can result in numerous negative consequences. In order to identify those who will benefit from a gluten free diet we need more studies to investigate specific biomarkers and epigenetics. More studies are needed to identify whether any other wheat component can be implied in the pathogenesis of inflammation and autoimmune diseases. Multiple enzyme replacement therapy have been tried recently with promising results in patients with gluten intolerance. Immune therapies are also being considered as adjuvant therapies owing to the role of immune system in the pathogenesis of NCGS [46].Trials have also shown the restoration of the beneficial gut microbiota with probiotic administration [48].

Key Words: *Non Celiac Gluten Sensitivity, Celiac disease, Gluten free diet*

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I. Introduction

Since the last few decades, global production and consumption of wheat have increased exponentially. Due to this, a wide variety of gluten-related disorders have surfaced and their prevalence has been increasing since then[1]. Gluten which consists of glutenin polymers and gliadin monomers is now being considered as "the new diet villain" [2]. There has been a recent widening of the spectrum of gluten-related disorders - celiac disease (CD), wheat allergy, and non-celiac gluten sensitivity (NCGS). These disorders are not clearly distinguishable and indeed overlap with each other. Even though the symptomology of these disorders are more or less the same, there is a vast diversity in the mechanisms that underlie their symptom generation. The lack of scientific evidence regarding the etiopathogenesis, epidemiology, and less reliable diagnostic criteria, makes it difficult to clearly understand and define these clinical conditions. The exact prevalence of NCGS is unknown, but it is now considered higher than CD. Only with a double blind placebo controlled (DPPC) gluten challenge diagnosis of NCGS can be confirmed. But this procedure has limited applicability in routine clinical practice due to its complexity. Differentiating NCGS from functional gastrointesyinal diseses like irritable bowel syndrome(IBS) is also a challenge because of the absence of reliable and reproducible diagnostic markers. The first reports on NCGS were published more than 40 years ago. Many researches are being done to identify the underlying mechanisms of NCGS to improve the diagnosis and treatment. New reports are being published in

this regard. In this literature review we discuss the up to date review on epidemiology, pathophysiology, diagnosis, differential diagnosis and treatment of NCGS.

II. Gluten-Related Disorders – What Are They?

Celiac Disease

Definition

Celiac disease is defined as a chronic small intestinal immune-mediated enteropathy precipitated by exposure to dietary gluten in genetically susceptible individuals. Duodenal mucosal biopsy shows crypt hypertrophy and villous atrophy. Normal histology is a strong negative predictor, especially in those on gluten-free diet.

Pathophysiology

The incompletely digested gliadin by its cytotoxic and immunomodulatory properties induce the enterocytes to produce interleukin (IL)15 and releases zonulin thereby increasing the gut permeability. The gluten peptides cross the intestinal epithelial barrier and reach the lamina propria where the glutamine residues are deaminated by the activated tissue transglutaminase (TG2). The glutamic acid thus formed interacts with the IL15 upregulated HLA class II on the antigen presenting cells. This triggers a cellular and humoral gliadin-specific immune response with the production of increased pro-inflammatory cytokines such as IFN-, tumor necrosis factor-alpha (TNF-) and IL-17 , which damage the intestinal mucosa[39]. The APCs present these antigens to CD4+ T lymphocytes in lamina propria which thereby gets activated. IL21 and interferon (IFN)γ released as a result of the activation in turn activates the intraepithelial CD8+ lymphocytes . This causes damage to enterocytes and cause villous atrophy. Cytokines released by activated T cells also contribute to the production of mainly IgA antibodies [37]. Inflammatory process may extend beyond the digestive system resulting in various extra intestinal manifestations like refractory iron deficiency anemia, dermatitis herpetiformis, ataxia,osteoporosis, delayed puberty, psychiatric manifestations etc. It may show autoimmune associations like diabetes mellites type 1, Addisons disease, IgA deficiency. It also shows associations with chromosomopathies like Down's syndrome and Turners syndrome. They have characteristic HLA DQ2 and/or HLA DQ8 halotypes, absence of which is a strong negative predictor of celiac disease.

Among the environmental factors, gastrointestinal dysbiosis seems to be associated to CD onset, consisting of an increased number of Proteobacteria and Bacteroidetes and a reduced number of Firmicutes, especially in the active phase of the disease.

Diagnosis is suspected by the presence of a suggestive clinical picture or a risk group. Serology shows positivity for immunoglobulin A (IgA) endomysial antibodies, IgA tissue transglutaminase antibodies, and IgG de-amidated gliadin peptide antibodies. These antibodies are usually of the IgA type and in cases of IgA deficiency; the same autoantibodies should be looked for in their IgG version. Duodenal biopsies obtained by endoscopy show villous atrophy/crypt hypertrophy/normal mucosal thickness with infiltrations in the lamina propria[38].

Wheat allergy, gluten intolerence and celiac disease are different conditions but with similar clinical presentations.In gluten intolerance there is intolerance to all grains containing gluten like wheat, rye, barley, and oats. Wheat allergy is allergic reactions to foods containing wheat and wheat varieties, not the other gluten-containing grains Those allergic to wheat are allergic to a part of wheat, either protein or non protein part, so allergy to wheat need not be always to its gluten content. All gluten-free foods are not wheat-free and not all wheat-free foods are gluten-free. Celiac disease is a chronic autoimmune disorder of the small intestine, with antibodies against the protein gluten. People with celiac disease do not get the fatal anaphylaxis when they eat wheat or gluten. But the immune system will eventually damage the small intestinal lining leading to malabsorption and severe malnutrition.

Irritable Bowel Syndrome

Definition

Irritable bowel syndrome (IBS) is a chronic functional bowel disorder and is one of the most commonly diagnosed gastrointestinal disorders worldwide with a global prevalence of about 10-20%.

Pathophysiology

The diagnosis of IBS is made based on Rome IV criteria. The patients present with long standing non specific symptoms like altered bowel habits, abdominal pain and absence of constitutional symptoms, weight loss, anorexia, gastrointestinal bleeding and fever. Majority of the report these symptoms following ingestion of one or more specific food groups. There are no biological markers available for diagnosis. IBS follows a multifactorial pathogenesis. Various mechanisms like visceral hypersensitivity, altered brain-intestine interactions, altered gut motility, gut microbiota, alterations in the intestinal permeability and bile acid

metabolism have been postulated. It is considered to be a biopsychosocial disease due to the role of the brain-intestine interactions in its development.

Non Celiac Gluten Sensitivity

Non celiac gluten sensitivity is a largely ambiguous group of disorders with evolving epidemiology. The increasing prevalence may be attributed to the increased wheat consumption over the last few decades. However the prevalence data is highly variable to lack of a proper well defined diagnostic criteria. Incidence is found to be more in young adult female population and in first degree relatives of CD [25][23][33].it is a non allergic and non autoimmune condition[43].The disorder is also known as gluten hypersensitivity or non-celiac gluten intolerance. This newly surfaced clinical entity has aroused so much interest in the scientific community.

The concept of a causal relationship between the ingestion of gluten and the occurrence of symptoms in absence of CD and wheat allergy was first described in the late 1970s by Cooper and Ellis. According to the Oslo definition, NCGS consists of "a variety of immunological, morphological, or symptomatic manifestations that are precipitated by the ingestion of gluten in individuals in whom celiac disease has been excluded"[5].

It is characterised by both intestinal and extra intestinal symptoms. This condition has also shown some neurological manifestations like cognitive dysfunction, memory disturbances, autism and so on [26].NCGS is considered as a subset of IBS as these conditions show a considerable clinical overlap. Both these can coexist independently without a common mechanism of pathogenesis [2]. Long term risk of development of complications is not considered with this disorder unlike CD.

Pathophysiology

Gluten, due to the presence of high concentrations of (20%) and glutamines (40%), is incompletely digested and produces peptides namely (33-mer)-gliadin fragment, cytotoxic peptide and interleukin releasing peptide which alter the gut permeability and produces disturbances in the mucosal innate immunity by its immunomodulatory effects. However the role of gluten as the cause of this condition is still uncertain. Based on a recent blinded randomised study, only a few cases showed recurrence of the symptoms after the blinded administration of gluten. This raises suspicion on the other components of wheat like alpha amylase- trypsin inhibitors [1][20], FODMAPs (fermentable, oligo-, di-, monosaccharides, and polyols)[15], fructans, lectins[30] and agglutinins[31]. Skodje and his colleagues found in his study that symptoms worsened in NCGS patients following consumption of fructan. These component in wheat causes increased gas production and osmotic effect in the gut [14][29]. Besides these triggers, other culprits for the same are still being investigated.Dietrich et al. studied the effect of a low FODMAPs versus a gluten free diet (GFD) on the symptomology, intestinal changes, microbiota changes and quality of life in NGCS patients. Biesiekierski et al. in his cohort studies also found a similar response to low FODMAPs[15]. Zanini et al.[16] and Elli et al.[17] did similar studies to analyse the role of FODMOPs in the pathogenesis of the condition but unfortunately were not able yield valid evidences.The findings pointed towards a multifactorial etiopathogenesis of this clinical condition which included immune response induced on exposure to gluten, microbiota shifts and functional effect of FODMAPs[15]. Hence "non-celiac wheat sensitivity" (NCWS) being a more inclusive term is now preferred to NCGS [42] .Here the patient experiences both gastrointestinal and extra-gastrointestinal symptoms with ingestion of gluten with no serological or histopathological evidence of celiac disease and wheat allergy. NCGS is also thought to cause psychiatric conditions like autism and schizophrenia. The study done in children with autism suggests that the symptoms may become more tolerable by a gluten-free diet[34].

The pathophysiology of NCGS is still unclear. Efforts have been made during the past few years to identify possible serological, immunological, histopathological, immunohistochemical and pathophysiological aspects characterizing this condition with the aim of using them for diagnostic purposes as demonstrated by the Consensus Conferences on NCGS held in London(2011),Munich(2012)and Salerno(2015) and by several scientific contributions. All these provide us with a largely heterogenous data.

Genomic studies

Very limited data is available regarding the genetics of NCGS. Patients with NCGS do not have any characteristic HLA haplotypes. About 50% of the patients have HLA DQ2 and HLA DQ8 but this is of little significance as this percentage is only midly elevated above the prevalence of these haplotypes in the general population[23]. Downregulation of the T-regulator marker FOXP3 was demonstrated indicating a possible role[12]. One study identified higher expression of 7 miRNA (hsa-miR-19b-3p, hsa-miR-19a-3p, hsa-miR-186-5p, hsa-miR 17-5p, hsa-miR-145-5p, hsa-miR-30e-5p, and hsa-miR-143-3p) in the intestinal mucosa of NCWS patients. These genes code for the mediators of inflammation and innate immunity. Genomic studies identified300 DEGs, of which around 64% were long non-coding RNA (lncRNA). These IncRNA may be important for the pathogenesis by processing and regulating the availability of mRNAs. Role of hedgehog signalling pathway and regulators of circadian rhythm have also been implicated in various studies [21][22] .

Sapone et al. [12] observed an up regulation in mRNA that codes for toll like receptors (TLR)-2 and claudin 4. TLRs are receptors activated by nonself-antigens during innate immune response. Claudin 4 is a member of a family of molecules which are integral components of tight junctions. However this observation did not warrant realiability in the successive studies. Picarelli et al. [19] found that immunohistochemical expression of TLR2 in duodenal mucosa of NCGS was comparable to that of CD and controls.

Immunohistochemistry

In NCGS, innate immune response to gluten peptides may be the predominant immune reaction responsible for the development of this condition[4]. The levels of CD14 lymphocytes and lipopolysaccharide binding protein [7] was found to be higher in NCGS when compared to healthy controls and CD. The presence of these biological markers which are indicators of innate immune activation against bacterial antigens confirms the hypothesized original pathogenetic pathway of this condition. The adaptive immune response has minimal role in NCGS unlike celiac disease. This explains the absence of the serological markers of CD in this condition. Nevertheless, Volta et al. in 2012 demonstrated positivity for IgG anti gliadin antibodies (found in CD) in about 56.4% cases and this was confirmed by successive cohort studies. However the serology became negative in 93.2% cases on adhering to gluten free diet. In another study carried out in Italy, 66% of patients with a diagnosis of NCGS showed the presence of anti-gliadin IgG antibodies (AGA).

Studies also speculate that the expression of interferon gamma transglutinase 2 in the intestinal mucosa in NCGS plays a role in its pathogenesis.

Immunohistochemistry studies[8][9]for CD4 (marker of T helper lymphocytes),CD8+ and CD117 were done. CD4 positivity was demonstrated with 100%s ensitivity and 90% specificity in differentiating CD from NCGS while 87.5% sensitivity and 85% specificity in differentiating healthy controls from NCGS. CD8 positive cell infiltration in the lamina propria was found to be intermediate between CD and healthy controls. Higher levels of CD117 positive cells were detected in NCGS.

It has been postulated that gliadin may induce mast cell degranulation which leads to inflammatory cytokine production resulting in a low grade intestinal inflammation.

Rectal biopsy samples obtained from patients with NCGS showed an increase in interferon gamma producing type-1 innate lymphoid cells with a characteristic CD45(+) T-bet(+), CD56(-), NKP44(-), and CD117(-) pattern which reverted by a gluten free diet[10].

Alterations in the gut

Some studies speculate the role of gut microbiota in the pathogenesis of the condition. The normal oral and intestinal microbiota produce gluten-degrading enzymes which might convert an immunogenic gluten-peptide to non-immunogenic peptide thereby leading to a cascade of events directed towards NCGS.

There are evidences which show alteration of intestinal permeability in NCGS. This dysfuction in the intestinal barrier is demonstrated in vivo, by lactulose-mannitol test by Sapone et al.[12] and zonulin assay (high zonulin serum levels demonstrated by Barbaro et al.[27]) while by analyzing TJ's protein expression, claudine-15 and myosin light chain kinase activity the same can be demonstrated ex vivo on intestinal biopsies.Hollon et al. analysed the intestinal permeability by measuring transepithelial electrical resistance in cultured duodenal biopsy samples[11] .He found out that gliadin increased intestinal permeability in NCGS more than in CD patients and healthy subjects.He also observed higher levels of fatty acid binding protein 2 which is a marker of epithelial damage and permeability in NCGS than in controls[7].Gluten is thought to increase the gut permeability and down regulate the expression of zona occludins in the intestinal mucosa resulting in dysfunctional intestinal barrier.

Histopathology and other postulations

Provocative gluten challenge test on immune cells was done in patients with NCGS both in vivo an in vitro. Increase in intraepithelial lymphocytes (CD3 staining) and mucosal interferon gamma with no change in heat shock protein 27 and 70 was observed in this cytokine analysis. Based on this observation, duodenal lymphocytosis i.e. more that 25 intraepithelial lymphocytes per 100 enterocytes was considered to be a risk factor for evolving into NCGS with an odds ratio of 28.59 .There was also an induced secretion of CXCL10 chemokine by peripheral mononuclear blood cells observed in these patients

Villanacci et al. conducted a single centre study and observed a linear T-lymphocyte infiltration (hallmarked by CD3 staining) in the lamina propria in about 78.5% of the patients included in the study. He proposed that this finding could characterize NCGS .

How to arrive at a NCGS diagnosis?

Kabbani et al devised an algorithm for the diagnosis of this new clinical entity which resulted in identifying CD in 7% of individuals with suspected NCGS while confirming NCGS in the remaining 93%. This

algorithm consisted of HLA typing, gluten challenge, serology and duodenal biopsy [32]. The current diagnostic criteria similar to this should include self-reported gluten intolerance with a negative serology for CD (including immunoglobulin A (IgA) endomysial antibodies, IgA tissue transglutaminase antibodies, and IgG de-amidated gliadin peptide antibodies)[4] and normal duodenal histology (Marsh 0 stage) as well as duodenal lymphocytosis with a normal villous architecture (Marsh 1 stage) whilst on a gluten-containing diet[13]. NCGS is distinguished from wheat allergy by the absence of IgE-wheat antibodies.

The diagnosis is confirmed once prompt response is seen with gluten exclusion and symptoms reappear with inclusion of gluten in the diet. "Salerno criteria" [6] is now being used to arrive at a diagnosis. The diagnostic protocol of this criterion involves an initial evaluation of a gluten free diet followed by a double-blind placebo-controlled challenge with crossover. Symptom evaluation is done with a Gastrointestinal Symptom Rating Scale /visual analogue scale during this phase. Double blind placebo controlled challenge with crossover is helpful in detecting the clinical condition but it requires a long duration (around 9 weeks),lacks practicability, has drawbacks of relying on subjective findings rather than objective findings and patient adherence may be questionable. To minimize the risk of positivity based simply on chance they have to repeated at least twice [4].

On applying gluten containing patches on the oral mucosa, if mucosal hyperemia, edema, blisters or burning occur in the mouth the test is said to be positive. The test showed statistically higher positive rate(75% of NCGS) than CD both under gluten containing (15%) and free diet (25%). This finding, when confirmed in a multicenter study involving a large population sample, could be used as a simple and effective test to diagnose NCGS.

There are no available biomarkers or other reliable diagnostic tests that will help to detect this disorder. Hence its diagnosis still remains a big challenge. Its detection is highly presumptive due to lack of scientific evidence and strong overlap with irritable bowel syndrome. It is therfore based on clinical exclusion.

It would be of great advantage if the biomarkers for this condition with no specific clinical contour are identified for diagnostic and treatment purposes[2][4].

Treatment

Gluten free diet and other non dietary interventions

Gluten free diet, as reported by the paediatrician Willem Karl Dicke[40], is being used since 1941 for the treatment of gluten related disorders.There is still confusion regarding whether to have a lifelong strict adherence to gluten free diet or have an "on demand" approach. Long term strict adherence to GFD can result in numerous negative consequences like macronutrient and micronutrient deficiencies, altered intestinal microbiota with reduction in beneficial flora. This also adds on to the patients financial burden. Negative social and psychological impacts have also been implicated due to the restrictive nature of the diet. Experts recommend reintroduction of gluten every 6 to 12 months along with periodic evaluation.

Biesiekierski, et al.[3]carried out a double-blind, placebo-controlled, re-challenge trial in Australia, on 34 IBS patients with diarrhoea as the predominant symptom. These patients who became symptomatically better after being on 6 weeks of gluten free diet are given either 16g of dietary gluten per day (19 patients) or gluten-free diet(15 patients)on a random basis. The symptomatic relief and level of general well being of the patients are assessed using a visual analogue scale. At the end of the study, a significantly greater number of patients in the gluten-containing group did not show improvement when compared to those on gluten free diet.(68% vs. 40%;p=0.001). However questions still remain unanswered regarding the sufficiency of elimination of dietary gluten alone for the control of symptoms.

Effects of gluten free diet

Even though a gluten free diet normalises the intestinal, biochemical and immunomodulatory changes that occur in the patients with NCGS, long term strict adherence to GFD comes with risks.

Due to lack of beneficial dietary components, both macronutrient as well as micronutrient deficiencies like deficiencies in vitamin D, iron, calcium, folate, riboflavin, niacin, thiamine and dietary fibers[41] have been observed in people on gluten free diet. This poses a serious threat to the general health and well being of the affected.

The normal gut microbiota populates the gastrointestinal tract in a craniocaudally increasing concentration gradient. They exhibit symbiosis with the host and helps in maintaining the gut integrity. Alterations in diet disrupts the normal composition. Studies conducted by De Palma et al.in 2009 and Hansen et al. analysed the changes in the gut microbiota in healthy individuals on GFD and observed decrease in commensal bacteria like Bifidobacterium, Clostridium lituseburense, Faecalibacteriumprausnitzii, E. hallii, A. Phaedrus, Dorea, Blautia and Lachnospiraceae[18] with a concomitant increase in the opportunistic pathogens like Enterobacteriaceae and Escherichia coli which can produce inflammatory changes in the gut mucosa. This

implies a detrimental effect of long-term strict adherence to GFD in both healthy individuals as well as NCGS patients. However, the effect of GFD on the gut microbiota still remains poorly elucidated.

Lebwohl et al. studied the development of coronary heart disease in individuals on GFD due to a lack of beneficiary whole grains in their diet. Heikkilä and colleagues were also in favour of this inference but lacked evidence to support the claim.

Potter and colleagues did a systematic review of 27 articles. He found increases but within the healthy range in high-density lipoproteins, fasting glucose levels, total cholesterol, and body mass index while no such finding was noticed with triglycerides, low-density lipoprotein, or blood pressure. The study thus concluded the association of GFD with coronary heart weak. However, this study also had limitations. Kim and his colleagues conducted a study which also supported this view. He also commented on the beneficial role of GFD in decreasing waist circumference.

Education about the potential risks is of great importance, especially to individuals who under the influence of social media adhere to the GFD without any diagnosis as these individuals do not require an immediate change in their diet. Though many studies have been conducted regarding the health complications of strict adherence to a gluten-free diet, the results are inconclusive and conflicting and hence demand further investigation.

A study was done during the year 2013-14 [35] to analyse the effects of short term reintroduction of gluten in a homogenous group of 24 NCGS diagnosed patients after obtaining written consent from the participants and ethics clearance. These candidates were put on a 3 week GFD. The adherence to the diet was evaluated with CDAT(Celiac Dietary Adherence Test). Those with high scores indicates poor adherence to the diet. After 3 weeks, dietary intervention in the form of low gluten diet (3.5–4 g gluten/day) for the 1st week, mid-gluten diet (6.7–8 g gluten/day) for the second week and high-gluten die(10–13 g gluten/day) for the third week is given. During this period the interventions were adjusted whenever the participant reported an adverse event. Following this the patient can continue his/her regular diet if favourable responses are observed. At the end of the study, it was concluded that the quality of life and well being(especially decreased emotional score as per SF 36 subscores) were significantly affected in the subgroup that received low gluten diet($p = 0.050$) while tolerance level could be noticed in the other two subgroups allowing some amount of gluten in their diet. The results suggest the possibility of inter-individual variability against gluten.

Other therapies

Multiple enzyme replacement therapy have been tried recently with promising results in patients with gluten intolerance. An exploratory clinical research in which an enzyme mixture of 4 enzymes(namely, leucine aminopeptidase derived from Aspergillus oryzae, semi alkaline protease derived from Aspergillus melleus, deuterolysin derived from Penicillium citrinum and cysteine protease derived from Carica papaya) in equal amounts that helps in the digestion of 33-mer peptide into ≤6-mer peptides was administered in a group of individuals with diagnosed NCGS. Following overnight fasting, a randomized, single-blind, placebo-controlled crossover study was done in which one group was administered two capsules containing 250 mg enzyme mixture thrice daily while the other group was given dextrin as placebo in the same pattern for 2 weeks along with gluten challenge. No significant differences in the serum levels of IL-8, TNF-α, or RANTES(regulated on activation, normal T cell expressed and secreted) were detected at the end of the study in any of the groups indicating the inclusion of only NCGS patients. CSI score was significantly lower in the group which received the enzyme mixture when compared to the group that received the placebo.[36].Another trial using AN-PEP, a prolyl peptidase derived from Aspergillus niger, also showed effectiveness in NCGS patients[44].Larazotide acetate,which restores the dyregulated intestinal permeability,could be used as adjuvant therapy used to tolerate minimal amounts of gluten in NCGS[45][46]. Immune therapies are also being considered as adjuvant therapies owing to the role of immune system in the pathogenesis of NCGS[46].De Angelis and his collaborators[47] proposed that a combination of probiotic strains can show effectiveness in gluten related disorders. Trials have also shown the restoration of the beneficial gut microbiota with probiotic administration [48].

III. Conclusion

NCGS is increasingly reported as the global wheat consumption has increases many times over the past few decades. This condition needs to be differentiated from wheat allergy ,celiac disease and functional gastrointestinal disorders as there is obvious overlap of symptoms. But there is lack of sensible and specific biomarkers for NCGS diagnosis making this process difficult. Although duodenal biopsy is useful to rule out a seronegative celiac disease it is not useful for a “positive diagnosis” of NCGS. So there is need for identification of reliable diagnostic tests which are accessible, available and affordable in routine clinical practice to improve the diagnosis and treatment of NCGS. Over the past ten years there is increasing awareness on the harmful effects of gluten and the number of reports on gluten free diets has been increasing in scientific journals and social media. This societal influence plays a major role in reshaping people’s food habit with many

turning to a wheat free diet. Since gluten free diet and a low FODMAP can lead to nutritional imbalance people should be encouraged to consult a clinical nutritionist before initiating such dietary restrictions. In order to identify those who will benefit from a gluten free diet we need more studies to investigate specific biomarkers and epigenetics. More studies are needed to identify whether any other wheat component can be implied in the pathogenesis of inflammation and autoimmune diseases.

References

[1] Husby S, Murray J. Non-Celiac Gluten Hypersensitivity: What Is All The Fuss About?. F1000prime Reports. 2015;7.

[2] Makharia A, Catassi C, Makharia GK. The Overlap Between Irritable Bowel Syndrome And Non-Celiac Gluten Sensitivity: A Clinical Dilemma. Nutrients. 2015 Dec 10;7(12):10417-26.

[3] Biesiekierski JR, Newnham ED, Irving PM, Barrett JS, Haines M, Doecke JD, Shepherd SJ, Muir JG, Gibson PR. Gluten Causes Gastrointestinal Symptoms In Subjects Without Celiac Disease: A Double-Blind Randomized Placebo-Controlled Trial. Official Journal Of The American College Of Gastroenterology| ACG. 2011 Mar 1;106(3):508-14.

[4] Ierardi E, Losurdo G, Piscitelli D, Giorgio F, Amoruso A, Iannone A, Principi M, Di Leo A. Biological Markers For Non-Celiac Gluten Sensitivity: A Question Awaiting For A Convincing Answer. Gastroenterology And Hepatology From Bed To Bench. 2018;11(3):203.

[5] Ludvigsson JF, Leffler DA, Bai JC, Biagi F, Fasano A, Green PH, Hadjivassiliou M, Kaukinen K, Kelly CP, Leonard JN, Lundin KE. The Oslo Definitions For Coeliac Disease And Related Terms. Gut. 2013 Jan 1;62(1):43-52.

[6] Catassi C, Elli L, Bonaz B, Bouma G, Carroccio A, Castillejo G, Cellier C, Cristofori F, De Magistris L, Dolinsek J, Dieterich W. Diagnosis Of Non-Celiac Gluten Sensitivity (NCGS): The Salerno Experts' Criteria. Nutrients. 2015 Jun 18;7(6):4966-77.

[7] Uhde M, Ajamian M, Caio G, De Giorgio R, Indart A, Green PH, Verna EC, Volta U, Alaedini A. Intestinal Cell Damage And Systemic Immune Activation In Individuals Reporting Sensitivity To Wheat In The Absence Of Coeliac Disease. Gut. 2016 Dec 1;65(12):1930-7.

[8] Brottveit M, Beitnes AC, Tollefsen S, Bratlie JE, Jahnsen FL, Johansen FE, Sollid LM, Lundin KE. Mucosal Cytokine Response After Short-Term Gluten Challenge In Celiac Disease And Non-Celiac Gluten Sensitivity. Official Journal Of The American College Of Gastroenterology| ACG. 2013 May 1;108(5):842-50.

[9] Alvisi P, De Fazio L, Valerii MC, Cavazza E, Salerno A, Lacorte D, Dinelli G, Spisni E. Responses Of Blood Mononucleated Cells And Clinical Outcome Of Non-Celiac Gluten Sensitive Pediatric Patients To Various Cereal Sources: A Pilot Study. International Journal Of Food Sciences And Nutrition. 2017 Nov 17;68(8):1005-12.

[10] Di Liberto D, Mansueto P, D'alcamo A, Pizzo ML, Presti EL, Geraci G, Fayer F, Guggino G, Iacono G, Dieli F, Carroccio A. Predominance Of Type 1 Innate Lymphoid Cells In The Rectal Mucosa Of Patients With Non-Celiac Wheat Sensitivity: Reversal After A Wheat-Free Diet. Clinical And Translational Gastroenterology. 2016 Jul 1;7(7):E178.

[11] Hollon J, Leonard Puppa E, Greenwald B, Goldberg E, Guerrerio A, Fasano A. Effect Of Gliadin On Permeability Of Intestinal Biopsy Explants From Celiac Disease Patients And Patients With Non-Celiac Gluten Sensitivity. Nutrients. 2015 Feb 27;7(3):1565-76.

[12] Sapone A, Lammers KM, Casolaro V, Cammarota M, Giuliano MT, De Rosa M, Stefanile R, Mazzarella G, Tolone C, Russo MI, Esposito P. Divergence Of Gut Permeability And Mucosal Immune Gene Expression In Two Gluten-Associated Conditions: Celiac Disease And Gluten Sensitivity. BMC Medicine. 2011 Dec;9:1-1.

[13] Ierardi E, Losurdo G, Iannone A, Piscitelli D, Amoruso A, Barone M, Principi M, Pisani A, Di Leo A. Lymphocytic Duodenitis Or Microscopic Enteritis And Gluten-Related Conditions: What Needs To Be Explored?. Annals Of Gastroenterology. 2017;30(4):380.

[14] Skodje GI, Sarna VK, Minelle IH, Rolfsen KL, Muir JG, Gibson PR, Veierød MB, Henriksen C, Lundin KE. Fructan, Rather Than Gluten, Induces Symptoms In Patients With Self-Reported Non-Celiac Gluten Sensitivity. Gastroenterology. 2018 Feb 1;154(3):529-39.

[15] Biesiekierski JR, Peters SL, Newnham ED, Rosella O, Muir JG, Gibson PR. No Effects Of Gluten In Patients With Self-Reported Non-Celiac Gluten Sensitivity After Dietary Reduction Of Fermentable, Poorly Absorbed, Short-Chain Carbohydrates. Gastroenterology. 2013 Aug 1;145(2):320-8.

[16] Zanini B, Baschè R, Ferraresi A, Ricci C, Lanzarotto F, Marullo M, Villanacci V, Hidalgo A, Lanzini A. Randomised Clinical Study: Gluten Challenge Induces Symptom Recurrence In Only A Minority Of Patients Who Meet Clinical Criteria For Non-Coeliac Gluten Sensitivity. Alimentary Pharmacology & Therapeutics. 2015 Oct;42(8):968-76.

[17] Elli L, Tomba C, Branchi F, Roncoroni L, Lombardo V, Bardella MT, Ferretti F, Conte D, Valiante F, Fini L, Forti E. Evidence For The Presence Of Non-Celiac Gluten Sensitivity In Patients With Functional Gastrointestinal Symptoms: Results From A Multicenter Randomized Double-Blind Placebo-Controlled Gluten Challenge. Nutrients. 2016 Feb 8;8(2):84.

[18] De Palma G, Nadal I, Collado MC, Sanz Y. Effects Of A Gluten-Free Diet On Gut Microbiota And Immune Function In Healthy Adult Human Subjects. British Journal Of Nutrition. 2009 Oct;102(8):1154-60.

[19] Picarelli A, Borghini R, Di Tola M, Marino M, Urciuoli C, Isonne C, Puzzono M, Porowska B, Rumi G, Lonardi S, Salemme M. Intestinal, Systemic, And Oral Gluten-Related Alterations In Patients With Nonceliac Gluten Sensitivity. Journal Of Clinical Gastroenterology. 2016 Nov 1;50(10):849-58.

[20] Junker Y, Zeissig S, Kim SJ, Barisani D, Wieser H, Leffler DA, Zevallos V, Libermann TA, Dillon S, Freitag TL, Kelly CP. Wheat Amylase Trypsin Inhibitors Drive Intestinal Inflammation Via Activation Of Toll-Like Receptor 4. Journal Of Experimental Medicine. 2012 Dec 17;209(13):2395-408.

[21] Clemente E, Efthymakis K, Carletti E, Capone V, Sperduti S, Bologna G, Marchisio M, Di Nicola M, Neri M, Sallese M. An Explorative Study Identifies Mirna Signatures For The Diagnosis Of Non-Celiac Wheat Sensitivity. Plos One. 2019 Dec 13;14(12):E0226478.

[22] Efthymakis K, Clemente E, Marchioni M, Di Nicola M, Neri M, Sallese M. An Exploratory Gene Expression Study Of The Intestinal Mucosa Of Patients With Non-Celiac Wheat Sensitivity. International Journal Of Molecular Sciences. 2020 Mar 13;21(6):1969.

[23] Sapone A, Bai JC, Ciacci C, Dolinsek J, Green PH, Hadjivassiliou M, Kaukinen K, Rostami K, Sanders DS, Schumann M, Ullrich R. Spectrum Of Gluten-Related Disorders: Consensus On New Nomenclature And Classification. BMC Medicine. 2012 Dec;10:1-2.

[24] Sallese M, Lopetuso LR, Efthymakis K, Neri M. Beyond The HLA Genes In Gluten-Related Disorders. Frontiers In Nutrition. 2020 Nov 12;7:575844.

[25] Volta U, Caio G, Tovoli F, De Giorgio R. Non-Celiac Gluten Sensitivity: An Emerging Syndrome With Many Unsettled Issues. Italian Journal Of Medicine. 2014;8(4):225-31.

[26] Ashat M, Kochhar R. Non-Celiac Gluten Hypersensitivity. Tropical Gastroenterology. 2015 May 7;35(2):71-8.

[27] Barbaro MR, Cremon C, Caio G, De Giorgio R, Volta U, Stanghellini V, Barbara G. Zonulin Serum Levels Are Increased In Non-Celiac Gluten Sensitivity And Irritable Bowel Syndrome With Diarrhea. Gastroenterology. 2015;148(4):S56-.

[28] Taraghikhah N, Ashtari S, Asri N, Shahbazkhani B, Al-Dulaimi D, Rostami-Nejad M, Rezaei-Tavirani M, Razzaghi MR, Zali MR. An Updated Overview Of Spectrum Of Gluten-Related Disorders: Clinical And Diagnostic Aspects. BMC Gastroenterology. 2020 Dec;20:1-2.

[29] Murray K, Wilkinson-Smith V, Hoad C, Costigan C, Cox E, Lam C, Marciani L, Gowland P, Spiller RC. Differential Effects Of Fodmaps (Fermentable Oligo-, Di-, Mono-Saccharides And Polyols) On Small And Large Intestinal Contents In Healthy Subjects Shown By MRI. Official Journal Of The American College Of Gastroenterology| ACG. 2014 Jan 1;109(1):110-9.

[30] Haas H, Falcone FH, Schramm G, Haisch K, Gibbs BF, Klaucke J, Pöppelmann M, Becker WM, Gabius HJ, Schlaak M. Dietary Lectins Can Induce In Vitro Release Of IL-4 And IL-13 From Human Basophils. European Journal Of Immunology. 1999 Mar;29(3):918-27.

[31] Dalla Pellegrina C, Perbellini O, Scupoli MT, Tomelleri C, Zanetti C, Zoccatelli G, Fusi M, Peruffo A, Rizzi C, Chignola R. Effects Of Wheat Germ Agglutinin On Human Gastrointestinal Epithelium: Insights From An Experimental Model Of Immune/Epithelial Cell Interaction. Toxicology And Applied Pharmacology. 2009 Jun 1;237(2):146-53.

[32] Aziz I, Lewis NR, Hadjivassiliou M, Winfield SN, Rugg N, Kelsall A, Newrick L, Sanders DS. A UK Study Assessing The Population Prevalence Of Self-Reported Gluten Sensitivity And Referral Characteristics To Secondary Care. European Journal Of Gastroenterology & Hepatology. 2014 Jan 1;26(1):33-9.

[33] Tanpowpong P, Broder-Fingert S, Katz AJ, Camargo Jr CA. Predictors Of Gluten Avoidance And Implementation Of A Gluten-Free Diet In Children And Adolescents Without Confirmed Celiac Disease. The Journal Of Pediatrics. 2012 Sep 1;161(3):471-5.

[34] Elder JH, Shankar M, Shuster J, Theriaque D, Burns S, Sherrill L. The Gluten-Free, Casein-Free Diet In Autism: Results Of A Preliminary Double Blind Clinical Trial. Journal Of Autism And Developmental Disorders. 2006 Apr;36:413-20.

[35] Roncoroni L, Bascuñán KA, Vecchi M, Doneda L, Bardella MT, Lombardo V, Scricciolo A, Branchi F, Elli L. Exposure To Different Amounts Of Dietary Gluten In Patients With Non-Celiac Gluten Sensitivity (NCGS): An Exploratory Study. Nutrients. 2019 Jan 10;11(1):136.

[36] Ido H, Matsubara H, Kuroda M, Takahashi A, Kojima Y, Koikeda S, Sasaki M. Combination Of Gluten-Digesting Enzymes Improved Symptoms Of Non-Celiac Gluten Sensitivity: A Randomized Single-Blind, Placebo-Controlled Crossover Study. Clinical And Translational Gastroenterology. 2018 Sep 1;9(9):E181.

[37] Lionetti E, Castellaneta S, Francavilla R, Pulvirenti A, Tonutti E, Amarri S, Barbato M, Barbera C, Barera G, Bellantoni A, Castellano E. Introduction Of Gluten, HLA Status, And The Risk Of Celiac Disease In Children. New England Journal Of Medicine. 2014 Oct 2;371(14):1295-303.

[38] Kelly CP, Bai JC, Liu E, Leffler DA. Advances In Diagnosis And Management Of Celiac Disease. Gastroenterology. 2015 May 1;148(6):1175-86.

[39] Garrote JA, Gómez-González E, Bernardo D, Arranz E, Chirdo F. Celiac Disease Pathogenesis: The Proinflammatory Cytokine Network. Journal Of Pediatric Gastroenterology And Nutrition. 2008 Aug;47:S27-32.

[40] Van Berge-Henegouwen GP, Mulder CJ. Pioneer In The Gluten Free Diet: Willem-Karel Dicke 1905-1962, Over 50 Years Of Gluten Free Diet. Gut. 1993 Nov;34(11):1473.

[41] Wild D, Robins GG, Burley VJ, Howdle PD. Evidence Of High Sugar Intake, And Low Fibre And Mineral Intake, In The Gluten-Free Diet. Alimentary Pharmacology & Therapeutics. 2010 Aug;32(4):573-81.

[42] Ferch CC, Chey WD. Irritable Bowel Syndrome And Gluten Sensitivity Without Celiac Disease: Separating The Wheat From The Chaff. Gastroenterology. 2012 Mar 1;142(3):664-6.

[43] Catassi C, Bai JC, Bonaz B, Bouma G, Calabrò A, Carroccio A, Castillejo G, Ciacci C, Cristofori F, Dolinsek J, Francavilla R. Non-Celiac Gluten Sensitivity: The New Frontier Of Gluten Related Disorders. Nutrients. 2013 Oct;5(10):3839-53.

[44] König J, Holster S, Bruins MJ, Brummer RJ. Randomized Clinical Trial: Effective Gluten Degradation By Aspergillus Niger-Derived Enzyme In A Complex Meal Setting. Scientific Reports. 2017 Oct 12;7(1):13100.

[45] Kelly CP, Green PH, Murray JA, Dimarino A, Colatrella A, Leffler DA, Alexander T, Arsenescu R, Leon F, Jiang JG, Arterburn LA. Larazotide Acetate In Patients With Coeliac Disease Undergoing A Gluten Challenge: A Randomised Placebo-Controlled Study. Alimentary Pharmacology & Therapeutics. 2013 Jan;37(2):252-62.

[46] Serena G, D'Avino P, Fasano A. Celiac Disease And Non-Celiac Wheat Sensitivity: State Of Art Of Non-Dietary Therapies. Frontiers In Nutrition. 2020 Sep 8;7:152.

[47] De Angelis M, Rizzello CG, Fasano A, Clemente MG, De Simone C, Silano M, De Vincenzi M, Losito I, Gobbetti M. VSL# 3 Probiotic Preparation Has The Capacity To Hydrolyze Gliadin Polypeptides Responsible For Celiac Sprue Probiotics And Gluten Intolerance. Biochimica Et Biophysica Acta (BBA)-Molecular Basis Of Disease. 2006 Jan 1;1762(1):80-93.

[48] Quagliariello A, Aloisio I, Bozzi Cionci N, Luiselli D, D'Auria G, Martinez-Priego L, Pérez-Villarroya D, Langerholc T, Primec M, Mičetić-Turk D, Di Gioia D. Effect Of Bifidobacterium Breve On The Intestinal Microbiota Of Coeliac Children On A Gluten Free Diet: A Pilot Study. Nutrients. 2016 Oct 22;8(10):660.