Annals of Gastroenterology (2025) **38,** 237-246

Brown bowel syndrome: a systematic review

Rena HM Cao[a], Jason Diab[b,c,d], Michael C. Grimm[c,e], Christophe R. Berney[b,c]

University of Sydney; Bankstown-Lidcombe Hospital, NSW; University of New South Wales, Sydney; University of Notre Dame, School of Medicine, Sydney; St George Hospital, Kogarah, NSW, Australia

Abstract

Brown bowel syndrome (BBS) is a rare disorder characterized by brown pigmentation of the intestinal wall, thought to be a consequence of lipofuscin accumulation. Celiac disease and vitamin E deficiency have been postulated to be risk factors. We systematically searched PubMed, Embase, Web of Science and Cochrane to identify all case reports and abstracts reporting clinical information on patients with a confirmed diagnosis of BBS. Forty-two studies met our inclusion criteria, including 63 patients with confirmed BBS. The most common symptoms of BBS were diarrhea (50.8%) and malnutrition (50.8%), followed by abdominal pain (39.7%) and vomiting (22.2%). BBS patients with celiac disease who presented with similar symptoms to non-celiac patients were significantly less likely to be hypoalbuminemic (15.4 vs. 45.5%) and showed a non-significant trend towards a higher mortality rate (36.4% vs. 15.4%). Nineteen (31.7%) BBS patients were also vitamin E deficient. The clinical presentation and outcomes in BBS patients with vitamin E deficiency and celiac disease were similar to those without vitamin E deficiency and celiac disease. Further studies are warranted to better define the diagnostic-therapeutic approach to patients with BBS.

Keywords Intestinal lipofuscinosis, ceroidosis, vitamin E deficiency

Ann Gastroenterol 2025; 38 (3): 237-246

Introduction

Brown bowel syndrome (BBS), also known as intestinal lipofuscinosis or intestinal ceroidosis, is a rare disorder classically characterized by a brown discoloration of the intestinal wall. The macroscopic brown appearance results from a pathologic accumulation of lipofuscin in the cytoplasm of smooth muscle cells of the *muscularis propria* and *muscularis mucosa* of the gastrointestinal tract. Lipofuscin is a yellow-brown lipid-containing granule formed as a waste product of oxidative metabolism, which accumulates in the lysosome of most cells with senescence. BBS was first reported by Pappenheimer and Victor, describing "ceroid" pigmentation in the intestinal musculature of 4 autopsy cases [1]. Whilst the exact pathogenesis of BBS remains unconfirmed, malnutrition from malabsorptive states, such as celiac disease, pancreatitis and post-gastrointestinal surgery (gastric bypass, bowel resection), has been thought to play a role [2-4]. It has been postulated that vitamin E deficiency is significant in contributing to lipofuscin accumulation as a result of oxidative stress to mitochondrial membranes in the relative absence of antioxidant protection. BBS most commonly manifests with non-specific symptoms, including diarrhea, weight loss, nausea and vomiting. Early diagnosis and commencement of medical management, including vitamin E supplementation, has been shown to be beneficial [5-7]. However, many cases have identified associated complications of intestinal dysmotility, such as small bowel dilatation, volvulus, intussusception and pseudo-obstruction, which often result in emergency surgical intervention [8,9].

To date, fewer than 70 cases have been formally documented, with no recent review of the disease. There is no current consensus on the epidemiology of this condition,

[a]Royal Prince Alfred Hospital, Camperdown, NSW, Australia; University of Sydney, Faculty of Medicine (Rena HM Cao); [b]Bankstown-Lidcombe Hospital, Bankstown, NSW, Australia (Jason Diab, Christophe R. Berney); [c]University of New South Wales, Faculty of Medicine and Health, Sydney (Jason Diab, Michael C. Grimm, Christophe R. Berney); [d]University of Notre Dame, School of Medicine, Sydney (Jason Diab); [e]University of New South Wales, Faculty of Medicine and Health, Sydney; St George Hospital, Kogarah, NSW, Australia (Michael C. Grimm)

Conflict of Interest: None

Correspondence to: Dr Rena Cao (BMed) (MD), Department of Gastroenterology, Royal Prince Alfred Hospital, 50 Missenden Road, Camperdown NSW 2050, Australia,
e-mail: rena.cao@health.nsw.gov.au

Received 22 December 2024; accepted 10 April 2025;
published online 28 April 2025

DOI: https://doi.org/10.20524/aog.2025.0965

based on existing literature databases, and a lack of general understanding concerning patient profile and outcomes. The objective of this systematic review was to outline the clinical profile of BBS, as well as investigations, management and outcomes of this rare disorder.

Methods

A combined automated and manual systematic database search was conducted using the electronic search engines PubMed, Embase, Cochrane, Web of Science and Google Scholar to identify relevant studies. The search used the keywords "brown bowel syndrome", "intestinal lipofuscinosis", and "intestinal ceroidosis" and was last conducted on 16 February, 2025. Reference lists of included studies were manually screened to identify additional relevant articles.

Inclusion criteria consisted of journal articles reporting on human participants (including children, adolescents, and adults) with a histopathologically confirmed diagnosis of BBS. The eligible study designs included randomized studies, case series, case reports, and conference abstracts. Studies were excluded if they were narrative or systematic reviews, animal studies, or histopathological reports without a corresponding case report.

Studies were independently screened by 2 reviewers (RC, JD) at both the title/abstract and full-text levels. They worked independently, and any discrepancies were resolved through discussion and, if necessary, by consulting a third reviewer (CB). No automation tools were used in the selection process.

Data extraction was performed by 2 independent reviewers using a predefined template. Extracted data included:

- Demographics (age, sex)
- Clinical symptoms and signs (diarrhea, abdominal pain, malnutrition, constipation, vomiting, abdominal distension, peripheral edema, bowel obstruction). Malnutrition was defined by cachexia or significant recent weight loss
- Previous medical and surgical history (alcohol abuse, celiac disease, pancreatitis, other gastrointestinal diseases, previous abdominal surgery)
- Investigations (fecal fat, serum vitamin E level (>11.6 μmol/L), serum vitamin D level (>50 nmol/L), serum albumin level (>35 g/L))
- Diagnostic investigations (colonoscopy, imaging modalities)
- Histopathological findings (lipofuscin-like granules in *muscularis propria* or *muscularis mucosa*)
- Surgical (biopsy, laparotomy, resection) and medical management (vitamin E treatment)
- Morbidity and in-hospital mortality

Additional extracted data included the presence of gastrointestinal comorbidities, such as ulcers (duodenal, pyloric), liver cirrhosis, diverticulosis, jejunal atresia, hepatomegaly/fatty liver, Crohn's disease, gastric adenocarcinoma, Whipple's disease, chronic jejunitis, unspecified malabsorption syndrome and exocrine pancreatic insufficiency. If data were missing or unclear, missing data were managed by assumption-based imputations where applicable, or the study was excluded from specific analyses.

Given that the included studies were primarily case reports and case series, the risk of bias was assessed based on key methodological considerations relevant to these study types. These included the clarity of patient demographics, the adequacy of case definitions, the completeness of clinical details, and the extent of follow-up information provided. Each study was evaluated by 2 independent reviewers, who worked separately to assess potential biases. Any discrepancies in judgment were resolved through discussion. No automation tools were used in the bias assessment process.

Statistical analysis

Statistical analysis was conducted using IBM SPSS Statistics version 26.0. Continuous variables were analyzed using mean, range and standard deviation. A comparison was performed between patients with and without celiac disease, given its potential role as a risk factor. Differences between categorical variables were analysed using Fisher's exact test, while Student's *t*-test was used for continuous variables. Mortality was assessed using univariate and multivariate logistic regression analysis. For the synthesis of results, studies were grouped based on clinical characteristics and management strategies. No meta-analysis was performed, given the heterogeneity in study designs and the lack of comparative data. However, sensitivity analyses were conducted to assess robustness by excluding studies with incomplete clinical data or a high risk of bias. Publication bias was not formally assessed in view of the descriptive nature of the included studies. The certainty of the evidence for each outcome was assessed narratively, considering study design limitations, potential sources of bias, and completeness of clinical data. The review adhered to the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) checklist to ensure methodological transparency and completeness (Supplementary Table 1) [10]. A flowchart detailing the study selection process is provided in Supplementary Fig 1.

Results

Study characteristics

A total of 42 studies across Europe, North America, Asia and Australia were included in the present review. Thirty-six were reports of a single case and 6 studies were case series. Nearly all studies reported data regarding clinical findings, histopathological diagnosis, and patients' outcomes. All studies included data on demographics (age, sex) as well as past medical or surgical history, and either medical or surgical management.

Patient characteristics

There were 64 patients with a confirmed diagnosis of BBS. The mean age was 54.1±18.9 years and patients were predominantly male (64.1%). Approximately one-third (28.4%) of the patients had a history of gastrointestinal disease, including gastrointestinal ulcers, Crohn's disease, Whipple's disease or unspecified malabsorption syndrome. Twenty-seven patients (40.3%) had previous abdominal surgery and 11 (16.4%) had a diagnosis of celiac disease. A history of bowel obstruction or pseudo-obstruction was noted in 9 (13.4%) patients (Table 1).

Table 1 Summary of clinical variables for brown bowel syndrome

Clinical variables	Value
Demographic data	
Age (years) mean±SD	54.1±18.9
Sex	
Male	41 (64.1%)
Female	23 (35.9%)
Symptoms and signs	
Diarrhea	33 (51.6%)
Abdominal pain	25 (37.3%)
Malnourishment	32 (48.5%)
Constipation	9 (20.9%)
Vomiting	14 (21.9%)
Abdominal distension	6 (9.0%)
Peripheral edema	11 (16.4%)
Bowel obstruction	3 (4.5%)
Past medical and surgical history	
Alcohol abuse	6 (9.0%)
Celiac disease	11 (16.4%)
Pancreatitis	3 (4.5%)
Other GIT disease	19 (28.4%)
Obstruction or pseudo-obstruction	9 (13.4%)
Previous abdominal surgery	27 (40.3%)
Biochemical investigations	
Elevated fecal fat (>7 g/24 h)	15 (22.4%)
Serum vitamin E deficiency <0.5 mg/dL	21 (31.3%)
Serum vitamin D deficiency <12 ng/mL	10 (14.9%)
Serum albumin deficiency <33 g/L	14 (20.9%)
Diagnostic investigations	
Colonoscopy	21 (31.3%)
X-ray	19 (28.4%)
Ultrasound	3 (4.5%)
Computed tomography	14 (20.9%)
Nil	30 (44.8%)
Histopathological findings	
Lipofuscin-like granules in *muscularis propria*	49 (73.1%)
Lipofuscin-like granules in *muscularis mucosa*	11 (16.4%)
Management	
Biopsy	22 (32.8%)
Laparotomy	47 (70.1%)
Resection	33 (49.3%)
Vitamin E treatment	19 (28.4%)
Morbidity	6 (9.0%)
Mortality	12 (17.9%)

SD, standard deviation; GIT, gastrointestinal tract; SD, standard deviation

Clinical presentation

Most symptoms were non-specific, with the most prevalent including diarrhea (51.6%), abdominal pain (37.3%), malnutrition (48.5%) and vomiting (21.9%). Less common symptoms included constipation (20.9%), peripheral edema (16.4%) and abdominal distension (9.0%, Table 1). There were no differences in the symptoms and signs between those with or without celiac disease (P>0.05, Table 2).

On presentation, approximately half the cohort (55.2%) had some form of imaging for investigation. The most common imaging modality was X-ray (28.4%), followed by computed tomography (20.9%) then ultrasound (4.5%). However, almost half the cohort (44.8%) did not receive any radiological investigation, and close to a third of patients (31.3%) received a colonoscopy as part of the diagnostic evaluation (Table 1).

Approximately one-third of the patients (31.3%) demonstrated low serum levels of vitamin E and 10 (14.9%) were found to have low serum levels of vitamin D.

Management

Most patients who presented with non-specific acute symptoms underwent laparotomy, which confirmed the diagnosis of BBS (70.1%); approximately a third of those procedures were for obstruction or pseudo-obstruction. Almost a third of the patients (28.4%) were treated conservatively with vitamin E replacement, and all patients who had surgery also received vitamin E replacement. BBS patients with celiac disease were more likely to present with hypoalbuminemia (45.5% vs. 16.1%, P=0.043) and showed a higher proportion of vitamin E deficiency compared to non-celiac patients, with a trend towards statistical significance (54.5% vs. 26.8%, P=0.086, Table 2). However, there was no statistically significant difference in the management of patients with celiac disease compared to those without (resection: 63.6% vs. 46.4%, P=0.340; vitamin E treatment: 36.4% vs. 26.8%, P=0.492). Patients with celiac disease and a diagnosis of BBS did not have higher odds of mortality compared to non-celiac disease patients, despite a statistical trend (36.4% vs. 14.3%, P=0.099, Table 2).

Histopathology

Histopathology was significantly more likely to demonstrate lipofuscin-like granules in the muscularis mucosa of BBS patients with celiac disease, compared to non-celiac (50% vs. 14.6%, P=0.040).

Discussion

First described in 1861 by German pathologist Doctor Ernst Wagner, the findings of BBS were reported as a brown

Table 2 Comparative analysis of clinical variables and outcomes patients with brown bowel syndrome, with or without celiac disease (*P<0.05)

Clinical variables	Celiac disease (n=11)	Without celiac disease (n=53)	P-value
Demographic data			
Age (years) mean±SD	56.5±12.3	53.6±20.1	0.652
Male	8 (72.7%)	33 (64.7%)	0.754
Female	3 (27.3%)	20 (35.3%)	
Symptoms and signs			
Diarrhea	5 (45.5%)	28 (52.8%)	0.747
Abdominal pain	3 (27.3%)	22 (39.3%)	0.518
Malnourishment	5 (45.5%)	27 (49.1%)	>0.99
Constipation	1 (9.1%)	8 (14.3%)	>0.99
Vomiting	0 (0.0%)	14 (25.0%)	0.103
Abdominal distension	1 (9.1%)	5 (8.9%)	>0.99
Peripheral edema	2 (18.2%)	9 (16.1%)	>0.99
Bowel obstruction	0 (0.0%)	3 (5.4%)	>0.99
Past medical and surgical history			
Alcohol abuse	1 (9.1%)	5 (8.9%)	>0.99
Pancreatitis	0 (0.0%)	3 (5.4%)	>0.99
Other GIT disease	1 (9.1%)	18 (32.1%)	0.159
Previous abdominal surgery	2 (18.2%)	25 (44.6%)	0.178
Obstruction or pseudo-obstruction	2 (18.2%)	7 (12.5%)	0.635
Biochemical investigations			
Elevated fecal fat (>7 g/24 h)	2 (18.2%)	13 (23.2%)	>0.99
Serum vitamin E <0.5 mg/dL	6 (54.5%)	15 (26.8%)	0.086
Serum vitamin D deficiency <12 ng/mL	1 (9.1%)	9 (16.1%)	>0.99
Serum albumin deficiency <33 g/L	5 (45.5%)	9 (16.1%)	0.043*
Diagnostic investigations		15 (26.8%)	
Colonoscopy	6 (54.5%)	16 (28.6%)	0.086
X-ray	3 (27.3%)	3 (5.4%)	>0.99
Ultrasound	0 (0.0%)	12 (21.4%)	>0.99
Computed tomography	2 (18.2%)	26 (46.4%)	>0.99
Nil	4 (36.4%)		0.742
Histopathological findings			
Lipofuscin-like granules in muscularis propria	8 (100.0%)	41 (80.4%)	0.329
Lipofuscin-like granules in muscularis mucosa	4 (50.0%)	7 (14.6%)	0.040*
Management			
Surgery			
Biopsy	5 (45.5%)	17 (30.4%)	0.483
Laparotomy	9 (81.8%)	38 (67.9%)	0.484
Resection	7 (63.6%)	26 (46.4%)	0.340
Vitamin E treatment	4 (36.4%)	15 (26.8%)	0.492
Morbidity	1 (9.1%)	5 (8.9%)	>0.99
Mortality	4 (36.4%)	8 (14.3%)	0.099

SD, standard deviation; GIT, gastrointestinal tract; SD, standard deviation

discoloration of human intestinal wall [8]. In 1946, Pappenheimer and Victor subsequently described the accumulation of "ceroid" in the smooth muscle cells of the small intestine and postulated the etiologic significance of vitamin E deficiency in BBS [1]. The pathologists observed an accumulation of a similar brown pigment in cirrhotic livers [11] and uterus [12] of rats maintained on a diet low in protein, fat and vitamin E. A century later, Toffler used the term "brown bowel syndrome" to describe a golden yellow pigment found within the muscle cells of the jejunal *muscularis* layer [9] (Fig. 1A [3]).

The incidence of BBS would appear to be extremely low, without any formal reporting to date. This could be a result of underdiagnosis, given that its largely non-specific symptoms are likely to contribute to a significant "pre-clinical" phase. Microscopically, BBS is characterized by the deposition of golden-brown granules, predominantly within the cytoplasm of smooth muscle cells of the *muscularis propria*, and occasionally in the *muscularis mucosae* and surrounding blood vessels, giving the intestines the typical gross appearance of "brown bowel" (Fig. 1B and C). Ceroid and lipofuscin are auto-fluorescent granules produced in human tissue as a result of oxidative stress [13]. The term "ceroid" is typically used to describe granules generated under pathological conditions such as malnutrition, hypoxia or infection [14], whereas

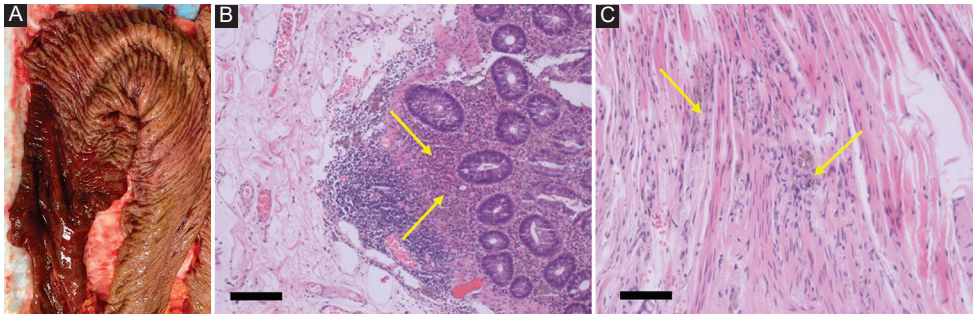


Figure 1 (A) Colonic specimen with clear transition point between normal bowel mucosa and diffuse dark brown pigmentation distally in a patient with brown bowel syndrome. (B) 10× magnification of submucosa and mucosa demonstrating brown pigmentation, scale bar = 1000 µm. (C) 40× magnification demonstrating pigmented macrophages on hematoxylin and eosin staining, scale bar = 250µm. Courtesy of Badiani [1], 2021, used with permission

lipofuscin refers to granules formed in aging post-mitotic cells as a result of the incomplete hydrolysis of oxidized lipids and proteins by lysosomal enzymes [15]. These terms are often used interchangeably in the literature to describe the histopathological findings of BBS, probably because of the indeterminate distinction on tissue sections. Many case reports have documented that these granules stain strongly positive for periodic-acid Schiff (PAS) stain [5,9,16]. Histologically, this distinguishes it from melanin, as it is only moderately positive for Fontana-Masson stain [4,5,16], and from iron [17-19], consistent with lipofuscin-like pigment.

Melanosis coli is a differential diagnosis to consider on endoscopic observation of bowel pigmentation. This is a reversible condition characterized by diffuse deposition of brown-black lipofuscin pigment in macrophages of the mucosal *lamina propria* and widely thought to be linked with chronic anthraquinone laxative use [20]. Although a well-recognized differential for BBS, laxative abuse was not reported in any of these patients.

In vitamin E deficiency, the pathogenesis of lipofuscin deposition in BBS is thought to be attributed to mitochondrial damage from excess oxidative stress. Vitamin E, which is lipid-soluble, has cellular anti-oxidant properties as a result of its ability to react with peroxyl radicals faster than the polyunsaturated fatty acids of the phospholipid membrane, thus exerting a protective effect on the mitochondrial membrane [17]. The phospholipid bilayer of mitochondrial membranes is exposed to oxidative damage by free radicals [18], leading to lipofuscin formation as a result of lysosomal degradation of the dysfunctional mitochondria [19]. The literature has reported numerous case studies of BBS with malabsorptive disorders, including celiac disease [21-29], following gastrointestinal surgery (bariatric, total gastrectomy), Crohn's, chronic idiopathic malabsorption [4,5,7,25,30-34] and pancreatitis [23,25,30,35]. In our study 40.3% of patients reported previous abdominal surgery, but more importantly, 16.4% suffered from celiac disease and 28.4% of patients had associated gastrointestinal pathologies, including gastrointestinal ulcers, Crohn's disease, Whipple's disease and diverticulosis. The correlation between BBS and intestinal dysmotility and subsequent obstruction or pseudo-obstruction has also been reported in several cases [3,5,16,25]. Our experience showed that a history of obstruction and pseudo-obstruction was indeed relevant, although it did not appear to be a common clinical presentation amongst the combined cohort (n=9, 13.4%, Table 1). One possible explanation is that it may have been underreported. Indeed, several articles included in our study still mentioned symptoms suggestive of either obstruction or pseudo-obstruction on presentation but did not formally record it. We believe that progressive disruption of muscle fiber architecture by lipofuscin accumulation in the myofibrils of the *muscularis propria* may ultimately lead to gut dysmotility and non-mechanical obstruction if left untreated [36].

BBS presents with a mixture of non-specific symptoms and signs, which renders its clinical diagnosis difficult in the absence of high clinical suspicion. The most common presenting symptoms were diarrhea, abdominal pain and malnutrition (Table 1). Abdominal pain was usually described as diffuse, ranging from colicky, to crampy or constant in nature. Only a small proportion of patients (20.9%) presented with constipation, which is surprising. We suspect this low incidence may be partially explained by a prolonged "pre-clinical" phase of this disease, followed by rapid transition to acute constipation and subsequent bowel obstruction, resulting in emergency surgery, often with a poor outcome.

The diagnosis of BBS relies upon transmural biopsy of the intestinal wall to capture the *muscularis propria*. Our study found that a significantly higher proportion of patients with vitamin E deficiency received full thickness intestinal biopsies, compared to patients without vitamin E deficiency (57.1% vs. 21.7%, P=0.010). One plausible explanation would be the association between lower serum vitamin E levels and advanced BBS, which in turn would be more likely to require surgical resection of the bowel. The majority (73.1%) of our cases identified lipofuscin-like granules accumulating in the *muscularis propria* and a smaller proportion (16.4%) distinctive accumulation in the *muscularis mucosa*. However, transmural biopsies are not commonly performed, compounding the challenges of diagnosis with the aforementioned factors.

Radiological evaluation with X-ray or computed tomography may be beneficial in identifying associated signs of obstruction or pseudo-obstruction. However, it is unlikely to contribute to the diagnosis of BBS. Conversely, colonoscopy may be a more useful diagnostic investigation tool, as BBS will often manifest

with gross intestinal wall discoloration. This said, mucosal changes might be too subtle to be recognized endoscopically. Only a third of patients underwent colonoscopies to investigate their presenting symptoms, most probably because of variations in the severity of their clinical findings. Indeed, up to one half of patients presented with acute signs of bowel obstruction, prompting exploratory laparotomy, where the diagnosis of BBS was ultimately made based on histopathological examination of the resected bowel, and therefore bypassing preoperative endoscopic investigation. Additionally, we identified that every case report that described a colonoscopy procedure was dated after 1979, suggesting that the low rate of colonoscopic investigation (31.3%) needs to be interpreted in the context of more limited availability of this diagnostic tool before 1979.

Several malabsorptive states can lead to a deficiency of fat-soluble vitamins A, D, E and K, including gastrointestinal causes, such as celiac disease [37-39], chronic pancreatitis [40] and Crohn's disease [41, 42], as well as post-surgical causes, such as gastrectomy [43, 44]. The impact of fat-soluble vitamin E deficiency has been reported widely, ranging from neurological manifestations, such as peripheral neuropathy or cerebellar dysfunction [45-48], to altered immune function in humans [49]. We demonstrated that vitamin E deficiency was observed in higher proportions in celiac patients compared to non-celiac with BBS (54.5% vs. 26.8%, P=0.086) (Table 2). With a trend towards significance, this may be attributed to the relatively small sample size, but also the increased recognition of asymptomatic celiac disease at an earlier stage throughout the last few decades. Indeed, the advent and widespread use of serological testing for celiac disease has resulted in a decreased percentage of patients presenting with gastrointestinal manifestations from underlying deficiencies and a higher proportion diagnosed with targeted screening [50]. We further demonstrated a significantly higher proportion of vitamin D deficiency in patients who were vitamin E deficient, compared with those who were vitamin E replete (33.3% vs. 6.5%, P=0.008) (Table 3). This may be accounted for by the malabsorptive or malnutritional state resulting in poor absorption of fat-soluble vitamins collectively. Furthermore, the impact of a chronic malabsorptive state showed celiac patients having significantly higher proportions of hypoalbuminemia compared to non-celiac BBS patients (45.5% vs. 16.1%, P=0.043) (Table 2).

The management of BBS depends on the severity of symptoms and associated complications on presentation. This, in turn, depends on the time of diagnosis. We reported that 70% of patients required laparotomy during their presentation, and that most of the cases published after year 2000 were for emergency indications, such as acute bowel obstruction, volvulus or pneumoperitoneum. In contrast, cases published before 2000 reported a significantly higher proportion of diagnostic exploratory laparotomies performed for non-urgent indications, mainly persistent abdominal pain and unexplained weight loss. This discrepancy is best explained by the increased availability of biochemical and advanced radiological investigation modalities in the 21st century, resulting in a reduction of unnecessary elective diagnostic laparotomy. Approximately one-fifth of our patients underwent a sub-total or total colectomy, including either end ileostomy, ileo-sigmoid or ileo-rectal anastomosis, with the extent of resection guided by the viability of the bowel at the time of diagnosis. The high proportion of emergency surgical intervention is probably attributed to delayed presentations, often resulting in acute bowel obstruction requiring imminent laparotomy and bowel resection [7,22,27,51,52].

In the case of incidental or early diagnosis of BBS, conservative medical management of the underlying cause of malabsorption, as well as vitamin E supplementation where necessary, may be sufficient to reverse symptoms [31,35,52,53]. In a recent case report of BBS, high-dose replacement of vitamin E at 268 mg twice daily resulted in significant clinical improvement [54]. Vitamin E supplementation has also been shown to reduce lipofuscin accumulation in fibroblasts derived from patients with neuromuscular degenerative disease [55], and murine brains and hearts [56,57]. Less than a third of our cohort were given vitamin E replacement therapy. This is probably an underestimate, as many case reports failed to indicate any specific medical management. Despite this, we still identified a significantly higher rate of vitamin E deficient patients receiving supplements as compared to the vitamin E replete group (47.6% vs. 19.6%, P=0.038). A plausible reason for the inconsistent administration of vitamin E therapy could simply be a failure to measure this critical biochemical parameter, due to incomplete understanding of the pathogenetic role of vitamin E in BBS. Our study indicated a wide variation in vitamin E dosage, ranging from 6 mg daily [22] to 750 mg daily [28], and no clear documentation of therapeutic duration. The lack of consensus on the therapeutic dose and duration outlines the paucity of clinical evidence supporting the benefit of vitamin E supplementation in BBS, mainly because of its extremely low incidence.

Neuronal ceroid lipofuscinosis, an inherited neurodegenerative lysosomal storage disorder, is another significant disease of lipofuscin accumulation. Emerging therapeutic studies in this area have targeted upstream disease mechanisms, including enzyme replacement therapy, gene therapy and stem cell therapy. In cellular models of neuronal ceroid lipofuscinosis disease, the potent antioxidant and nucleophilic agents phosphocysteamine and N-acetylcysteine showed activity in reducing ceroid accumulation [58]. A small follow-up clinical study demonstrated some reduction in ceroid deposits [59], although the clinical significance of this is unclear and it is unlikely to be applicable to BBS patients, as the therapy targets a different upstream mechanism of lipofuscinosis. Recently, research into Stargardt disease (inherited retinal disease of lipofuscin accumulation) and age-related macular degeneration has discovered a promising ability of the molecule soraprazan (renamed Remofuscin) in lipofuscin from retinal pigment epithelium [60-62]. *In vitro* and *in vivo* studies in human and murine retinal pigment epithelium cells respectively have shown a reduction of lipofuscin accumulation after treatment with Remofuscin, possibly secondary to generation of reactive oxygen species, specifically superoxide [63]. In murine models of Stargardt disease, supplementation of omega-3 fatty acids reduced lipofuscin accumulation [64].

Table 3 Comparative analysis of clinical variables and outcomes in patients with brown bowel syndrome, with or without vitamin E deficiency (*P<0.05)

Clinical variables	Vitamin E deficiency (n=21)	No vitamin E deficiency (n=43)	P-value
Demographic data			
Age (years) mean±SD	49.1±18.3	56.1±19.1	0.220
Male	17 (81.0%)	24 (55.8%)	0.040*
Female	4 (19.0%)	19 (44.2%)	
Symptoms and signs			
Diarrhea	13 (61.9%)	20 (46.5%)	0.294
Abdominal pain	7 (33.3%)	18 (39.1%)	0.787
Malnourishment	12 (60.0%)	20 (43.5%)	0.286
Constipation	2 (9.5%)	7 (15.2%)	0.709
Vomiting	6 (28.6%)	8 (17.4%)	0.340
Abdominal distension	1 (4.8%)	5 (10.9%)	0.657
Peripheral edema	4 (19.0%)	7 (15.2%)	0.730
Bowel obstruction	1 (2.2%)	2 (9.5%)	0.229
Past medical and surgical history			
Alcohol abuse	4 (19.0%)	2 (4.3%)	0.072
Pancreatitis	2 (9.5%)	1 (2.2%)	0.229
Celiac disease	6 (28.6%)	5 (10.9%)	0.086
Other GIT disease	4 (19.0%)	15 (32.6%)	0.382
Previous abdominal surgery	10 (47.6%)	17 (37.0%)	0.433
Obstruction or pseudo-obstruction	4 (19.0%)	5 (10.9%)	0.446
Biochemical investigations			
Elevated fecal fat (>7 g/24 h)	7 (33.3%)	8 (17.4%)	0.207
Serum vitamin D deficiency <12 ng/mL	7 (33.3%)	3 (6.5%)	0.008*
Serum albumin deficiency <33 g/L	6 (28.6%)	8 (17.4%)	0.340
Diagnostic investigations			
Colonoscopy	8 (38.1%)	13 (28.3%)	0.571
X-ray	10 (47.6%)	9 (19.6%)	0.038*
Ultrasound	0 (0.0%)	3 (6.5%)	0.546
Computed tomography	3 (14.3%)	11 (23.9%)	0.522
Nil	10 (47.6%)	20 (43.5%)	0.796
Histopathological findings			
Lipofuscin-like granules in muscularis propria	13 (83.7%)	36 (81.3%)	>0.99
Lipofuscin-like granules in muscularis mucosa	5 (31.3%)	6 (15.0%)	0.263
Management			
- Biopsy	12 (57.1%)	10 (21.7%)	0.010*
- Laparotomy	16 (76.2%)	31 (67.4%)	0.571
- Resection	9 (52.2%)	24 (42.9%)	0.600
Vitamin E treatment	10 (47.6%)	9 (19.6%)	0.038*
Morbidity	1 (4.8%)	5 (10.9%)	0.657
Mortality	1 (4.8%)	11 (23.9%)	0.086

SD, standard deviation; GIT, gastrointestinal tract; SD, standard deviation

Autophagy is a lysosome-dependent cellular mechanism for degrading unnecessary or dysfunctional components. Some studies have observed reduced levels of autophagy in senescent cells, and therefore suggested a role for autophagy enhancement in reducing lipofuscin accumulation. A number of agents have been shown to reduce lipofuscin accumulation, including anti-helminthic flubendazole [65] and zinc [66] in human retinal pigment epithelium, rapamycin in animal cardiomyocytes [67] and murine hepatocytes [68]. Although these results have been focused on extraintestinal cells, they present potential options for further therapeutic research in BBS. Ultimately, however, applying a traditional drug discovery approach (disease mechanism to target to therapy) to BBS is challenging, because of our limited understanding of its pathogenesis.

We identified a higher mortality rate in celiac patients with BBS, compared with non-celiac (36.4% vs. 14.3%, P=0.099), although the difference was not statistically significant (Table 2). This corroborates previous reports showing that patients with celiac disease have higher all-cause mortality rates [69,70]. Surprisingly, there was a trend towards a higher mortality rate in non-vitamin E deficient patients compared to vitamin E deficient patients (23.90% vs. 4.8%, P=0.086, Table 3). One possible explanation is that, although vitamin E deficiency is a likely precursor for the development of BBS,

other comorbidity factors may have played a more significant role with regard to the poorer outcomes. Unfortunately, those extended data were lacking.

One major limitation of this study is the incompleteness and inconsistency of the data presented across the case reports, probably because of the rarity of the disease. Given the very low number of cases of BBS documented in the literature, our inclusion criteria encompassed case reports dating back as early as 1946, as well as abstracts, adding heterogeneity of management and diagnostic criteria, as well as details of patient presentation, biochemical investigations and histopathology findings. The evolving availability of such investigations, imaging modalities and knowledge about this disease over the last 75 years partially accounts for the varying level of clinical details included in those case reports. Recent studies generally had broader information with respect to biochemical, endoscopic and radiological investigations. The wide historical span of cases included in our study also captured some of the changing diagnostic and management practices. This evolving availability of technology and advanced investigation modalities poses potential confounding variables for results, including the proportions of patients undergoing colonoscopy or laparotomy, respectively. Additionally, the extremely low incidence of this disease and the paucity of evidence concerning confirmed risk factors for BBS required us to make an informed decision in selecting which variables might be more relevant when comparing patient groups. We chose vitamin E deficiency, as this was understood to be a significant contributor to the pathogenesis of BBS, and celiac disease status, as this was indicated to be the primary diagnosis in several BBS cases [51], based on our literature

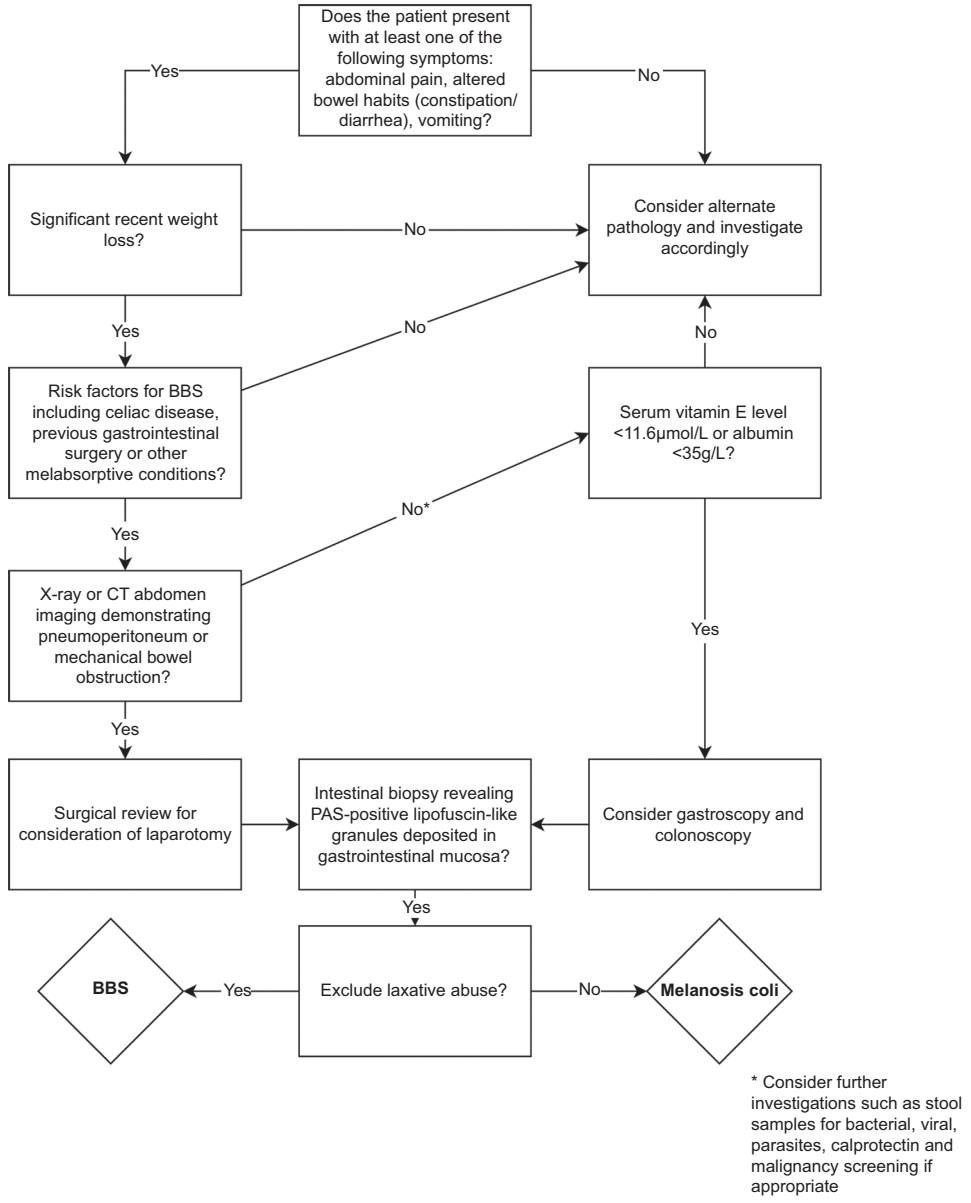


Figure 2 Clinical algorithm for diagnosis of brown bowel syndrome (BBS)

search. Finally, another limitation is the absence of a definitive cutoff for vitamin E levels that could potentially help with the future development of diagnostic criteria for BBS, and the lack of any specific biochemical diagnostic markers to date. This is a clear limitation, both for our study, and also for the standardized diagnosis of BBS. This said, we have created a clinical algorithm for the diagnosis and management of BBS that may help our readers: it is summarized in Fig. 2.

Concluding remarks

BBS is a rare phenomenon characterized by pathological accumulation of lipofuscin in the *muscularis* layer of the intestinal wall. BBS is not a primary condition; it is probably a complication resulting from chronic vitamin E deficiency, usually in the context of malnutrition or malabsorption. Currently there are no defined diagnostic parameters for BBS, including vitamin E cutoff levels. This probably contributes to delayed presentations and the development of associated complications, including gut dysmotility and pseudo-obstruction, requiring acute surgical intervention. Management should focus on treating the underlying cause of malnutrition or malabsorption where possible, but replacement of deficient fat-soluble vitamins should be commenced in a timely manner. Our study demonstrated that there was no significant difference in the incidence of vitamin E deficiency and celiac disease in patients with BBS. Further investigations are warranted into the risk factors for BBS, clearer diagnostic parameters and the effectiveness of vitamin E replacement therapy in reversing histopathological BBS.

References

1. Pappenheimer AM, Victor J. Ceroid pigment in human tissues. *Am J Pathol* 1946;**22**:395-413.
2. Arnold CA, Burke AP, Calomeni E, et al. Brown bowel syndrome: a multi-institutional case series. *Am J Surg Pathol* 2020;**44**:834-837.
3. Badiani S, Diab J, Chou R, Berney CR. Brown bowel syndrome: a rare cause of intestinal obstruction. *ANZ J Surg* 2021;**91**:2205-2207.
4. Lee H, Carlin AM, Ormsby AH, Lee MW. Brown bowel syndrome secondary to jejunoileal bypass: the first case report. *Obes Surg* 2009;**19**:1176-1179.
5. Ward HC, Leake J, Milla PJ, Spitz L. Brown bowel syndrome: a late complication of intestinal atresia. *J Pediatr Surg* 1992;**27**: 1593-1595.
6. Lee SP, Nicholson GI. Ceroid enteropathy and vitamin E deficiency. *N Z Med J* 1976;**83**:318-320.
7. Parente R, Pinamonti M, Martina S. Brown bowel syndrome in a middle-aged woman with chronic idiopathic malabsorption. *Case Rep Surg* 2019;**2019**:4706592.
8. Wagner E. Uber eine eigenthumliche primare Fettmetamorphose der Muskelhaut des Dunndarms. *Arch Heilkunde* 1861;**2**:455-459.
9. Toffler AH, Hukill PB, Spiro HM. Brown bowel syndrome. *Ann Intern Med* 1963;**58**:872-877.
10. Page MJ, McKenzie JE, Bossuyt PM, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. *BMJ* 2021;**372**:n71.
11. Lillie RD, Daft FS, Sebrell WH. Cirrhosis of the liver in rats on a deficient diet and the effect of alcohol. *Public Health Reports (1896-1970)* 1941;**56**:1255-1258.
12. Barrie MM. Vitamin E deficiency in the rat: fertility in the female. *Biochem J* 1938;**32**:2134-2137.
13. Sohal RS, Brunk UT. Lipofuscin as an indicator of oxidative stress and aging. *Adv Exp Med Biol* 1989;**266**:17-26.
14. Porta EA. Pigments in aging: an overview. *Ann N Y Acad Sci* 2002;**959**:57-65.
15. Brunk UT, Terman A. Lipofuscin: mechanisms of age-related accumulation and influence on cell function. *Free Radic Biol Med* 2002;**33**:611-619.
16. Ponce-Zepeda J, Guo W, Carmen G, et al. Brown bowel syndrome is a rare and commonly missed disease: a case report and literature review. *Case Rep Gastrointest Med* 2021;**2021**:6684678.
17. Miyazawa T, Burdeos GC, Itaya M, Nakagawa K, Miyazawa T. Vitamin E: regulatory redox interactions. *IUBMB Life* 2019;**71**:430-441.
18. Bhatti JS, Bhatti GK, Reddy PH. Mitochondrial dysfunction and oxidative stress in metabolic disorders—a step towards mitochondria based therapeutic strategies. *Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease* 2017;**1863**:1066-1077.
19. König J, Ott C, Hugo M, et al. Mitochondrial contribution to lipofuscin formation. *Redox Biol* 2017;**11**:673-681.
20. Yang N, Ruan M, Jin S. Melanosis coli: a comprehensive review. *Gastroenterol Hepatol* 2020;**43**:266-272.
21. Waldron D, Horgan P, Barry K, et al. Anorectal functional deficit in the brown bowel syndrome. *Ir J Med Sci* 1994;**163**:404-405.
22. Robinson MH, Dowling BL, Clark JV, Mason CH. Brown bowel syndrome: an unusual cause of massive dilatation of the colon. *Gut* 1989;**30**:882-884.
23. Reynaert H, Debeuckelaere S, De Waele B, et al. The brown bowel syndrome and gastrointestinal adenocarcinoma. Two complications of vitamin E deficiency in celiac sprue and chronic pancreatitis? *J Clin Gastroenterol* 1993;**16**:48-51.
24. Musumba C, Campbell F, Leiper K. Acute abdomen in a woman with celiac disease. *Gastroenterology* 2011;**141**:e11-e12.
25. Stamp GW, Evans DJ. Accumulation of ceroid in smooth muscle indicates severe malabsorption and vitamin E deficiency. *J Clin Pathol* 1987;**40**:798-802.
26. Hurley JP, Leary R, Connolly CE, Keeling P. Massive lower gastrointestinal bleeding in association with the brown bowel syndrome. *J R Soc Med* 1991;**84**:437-438.
27. Alwatari Y, Preciado C, Tondon R, Shiraff A. Brown bowel syndrome presenting as a small bowel obstruction. *AJSP Rev Rep* 2018;**23**:262-265.
28. Ruchti C, Eisele S, Kaufmann M. Fatal intestinal pseudo-obstruction in brown bowel syndrome. *Arch Pathol Lab Med* 1990;**114**:76-80.
29. Reyes Martínez C, Reina Campos FR, García Fernández FJ, et al. [Brown bowel syndrome]. *Rev Esp Enferm Dig* 2001;**93**:331-332.
30. Fullerton PM. Pigmentation of jejunal muscle. *Br Med J* 1960;**1**: 249-251.
31. Michowitz M, Noy S, Chayen D, Baratz M, Bawnik JB. Brown-bowel syndrome. *Am Surg* 1989;**55**:566-569.
32. Lee SA, Kim HK, Bae JY, et al. Brown bowel syndrome that developed after total gastrectomy - a case report. *Korean J Pathol* 2008;**42**:165-168.
33. Lambert JR, Luk SC, Pritzker KP. Brown bowel syndrome in Crohn's disease. *Arch Pathol Lab Med* 1980;**104**:201-205.
34. Soares PFDC, de Carvalho RB, Chaim EA, Cazzo E. Brown bowel syndrome: a rare malnutrition-related complication of bariatric surgery. *Nutr Hosp* 2019;**36**:743-747.
35. Bauman MB, DiMase JD, Oski F, Senior JR. Brown bowel and skeletal myopathy associated with vitamin E depletion in pancreatic insufficiency. *Gastroenterology* 1968;**54**:93-100.

36. Feeney EJ, Austin S, Chien YH, et al. The value of muscle biopsies in Pompe disease: identifying lipofuscin inclusions in juvenile- and adult-onset patients. *Acta Neuropathol Commun* 2014;**2**:2.
37. McGrogan L, Mackinder M, Stefanowicz F, et al. Micronutrient deficiencies in children with coeliac disease; a double-edged sword of both untreated disease and treatment with gluten-free diet. *Clin Nutr* 2021;**40**:2784-2790.
38. Hozyasz KK, Chelchowska M, Laskowska-Klita T. [Vitamin E levels in patients with celiac disease]. *Med Wieku Rozwoj* 2003;**7**:593-604.
39. Lodhi MU, Stammann T, Kuzel AR, Syed IA, Ishtiaq R, Rahim M. Celiac disease and concomitant conditions: a case-based review. *Cureus* 2018;**10**:e2143.
40. Martínez-Moneo E, Stigliano S, Hedström A, et al. Deficiency of fat-soluble vitamins in chronic pancreatitis: A systematic review and meta-analysis. *Pancreatology* 2016;**16**:988-994.
41. Fabisiak N, Fabisiak A, Watala C, Fichna J. Fat-soluble vitamin deficiencies and inflammatory bowel disease: systematic review and meta-analysis. *J Clin Gastroenterol* 2017;**51**:878-889.
42. Hwang C, Ross V, Mahadevan U. Micronutrient deficiencies in inflammatory bowel disease: from A to zinc. *Inflamm Bowel Dis* 2012;**18**:1961-1981.
43. Rino Y, Oshima T, Yoshikawa T. Changes in fat-soluble vitamin levels after gastrectomy for gastric cancer. *Surg Today* 2017;**47**:145-150.
44. Bloomberg RD, Fleishman A, Nalle JE, Herron DM, Kini S. Nutritional deficiencies following bariatric surgery: what have we learned? *Obes Surg* 2005;**15**:145-154.
45. Battisti C, Dotti MT, Formichi P, et al. Disappearance of skin lipofuscin storage and marked clinical improvement in adult onset coeliac disease and severe vitamin E deficiency after chronic vitamin E megatherapy. *J Submicrosc Cytol Pathol* 1996;**28**:339-344.
46. Suárez Llanos JP, González Melo E, Mora Mendoza A, Iacampo Leiva LD, Moro Miguel MA. Celiac disease and malabsorption: a case report of ataxia secondary to vitamin E deficiency. *Endocrinol Nutr* 2014;**61**:389-390.
47. Mauro A, Orsi L, Mortara P, Costa P, Schiffer D. Cerebellar syndrome in adult celiac disease with vitamin E deficiency. *Acta Neurol Scand* 1991;**84**:167-170.
48. Ulatowski LM, Manor D. Vitamin E and neurodegeneration. *Neurobiol Dis* 2015;**84**:78-83.
49. Pekmezci D. Chapter eight - vitamin E and immunity. In: Litwack G, ed. Vitamins & Hormones. Volume 86: Academic Press, 2011:179-215.
50. Ravikumara M, Tuthill DP, Jenkins HR. The changing clinical presentation of coeliac disease. *Arch Dis Child* 2006;**91**:969-971.
51. Białas M, Demczuk S, Dyduch G, Drabik G, Chrupek M, Okoń K. Brown bowel syndrome (intestinal lipofuscinosis) - a case report and review of the literature. *Pol J Pathol* 2013;**64**:228-231.
52. Chen Wongworawat Y, Propst R, Raza A, et al. Severe chronic bowel obstruction associated with brown bowel syndrome. *SAGE Open Med Case Rep* 2020;**8**:2050313X20945531.
53. Hosler JP, Kimmel KK, Moeller DD. The "brown bowel syndrome": a case report. *Am J Gastroenterol* 1982;**77**:854-855.
54. Alkurdi A, Rubin D, Seelhoff A, Herbst H. Brown bowel syndrome: an exceedingly rare condition with longstanding malabsorption and an unusual cause of colon pseudo-obstruction. *Case Rep Gastroenterol* 2021;**15**:960-965.
55. Villalón-García I, Álvarez-Córdoba M, Povea-Cabello S, et al. Vitamin E prevents lipid peroxidation and iron accumulation in PLA2G6-Associated Neurodegeneration. *Neurobiol Dis* 2022;**165**:105649.
56. Monji A, Morimoto N, Okuyama I, Yamashita N, Tashiro N. Effect of dietary vitamin E on lipofuscin accumulation with age in the rat brain. *Brain Res* 1994;**634**:62-68.
57. Ma XY, Su YB, Zhang FR, Li JF. Effects of vitamin E on the blastogenic response of splenocytes and lipofuscin contents in the hearts and brains of aged mice. *J Environ Pathol Toxicol Oncol* 1996;**15**:51-53.
58. Zhang Z, Butler JD, Levin SW, Wisniewski KE, Brooks SS, Mukherjee AB. Lysosomal ceroid depletion by drugs: therapeutic implications for a hereditary neurodegenerative disease of childhood. *Nat Med* 2001;**7**:478-484.
59. Levin SW, Baker EH, Zein WM, et al. Oral cysteamine bitartrate and N-acetylcysteine for patients with infantile neuronal ceroid lipofuscinosis: a pilot study. *Lancet Neurol* 2014;**13**:777-787.
60. Julien S, Schraermeyer U. Lipofuscin can be eliminated from the retinal pigment epithelium of monkeys. *Neurobiol Aging* 2012;**33**:2390-2397.
61. Taubitz T, Peters T, Pöschel S, et al. Removal of lipofuscin from the RPE of Abca4-/- mice with THPE: quantitative and toxicity studies. *Invest Ophthalmol Vis Sci* 2015;**56**:4199.
62. Dhooge P, Möller P, Boon C, et al. The STArgardt Remofuscin Treatment Trial (STARTT): design and baseline characteristics of enrolled Stargardt patients. *Open Res Europe* 2022;**1**:96.
63. Fang Y, Taubitz T, Tschulakow AV, et al. Removal of RPE lipofuscin results in rescue from retinal degeneration in a mouse model of advanced Stargardt disease: role of reactive oxygen species. *Free Radic Biol Med* 2022;**182**:132-149.
64. Prokopiou E, Kolovos P, Kalogerou M, et al. Omega-3 fatty acids supplementation: therapeutic potential in a mouse model of Stargardt disease. *Invest Ophthalmol Vis Sci* 2018;**59**:2757-2767.
65. Zhang Q, Presswalla F, Ali RR, Zacks DN, Thompson DA, Miller JML. Pharmacologic activation of autophagy without direct mTOR inhibition as a therapeutic strategy for treating dry macular degeneration. *Aging (Albany NY)* 2021;**13**:10866-10890.
66. Blasiak J, Pawlowska E, Chojnacki J, Szczepanska J, Chojnacki C, Kaarniranta K. Zinc and autophagy in age-related macular degeneration. *Int J Mol Sci* 2020;**21**:4994.
67. Li WW, Wang HJ, Tan YZ, Wang YL, Yu SN, Li ZH. Reducing lipofuscin accumulation and cardiomyocytic senescence of aging heart by enhancing autophagy. *Exp Cell Res* 2021;**403**:112585.
68. Martínez-Cisuelo V, Gómez J, García-Junceda I, et al. Rapamycin reverses age-related increases in mitochondrial ROS production at complex I, oxidative stress, accumulation of mtDNA fragments inside nuclear DNA, and lipofuscin level, and increases autophagy, in the liver of middle-aged mice. *Exp Gerontol* 2016;**83**:130-138.
69. West J, Logan RFA, Smith CJ, Hubbard RB, Card TR. Malignancy and mortality in people with coeliac disease: population based cohort study. *BMJ* 2004;**329**:716-719.
70. Anderson LA, McMillan SA, Watson RG, et al. Malignancy and mortality in a population-based cohort of patients with coeliac disease or "gluten sensitivity". *World J Gastroenterol* 2007;**13**:146-151.

Supplementary material

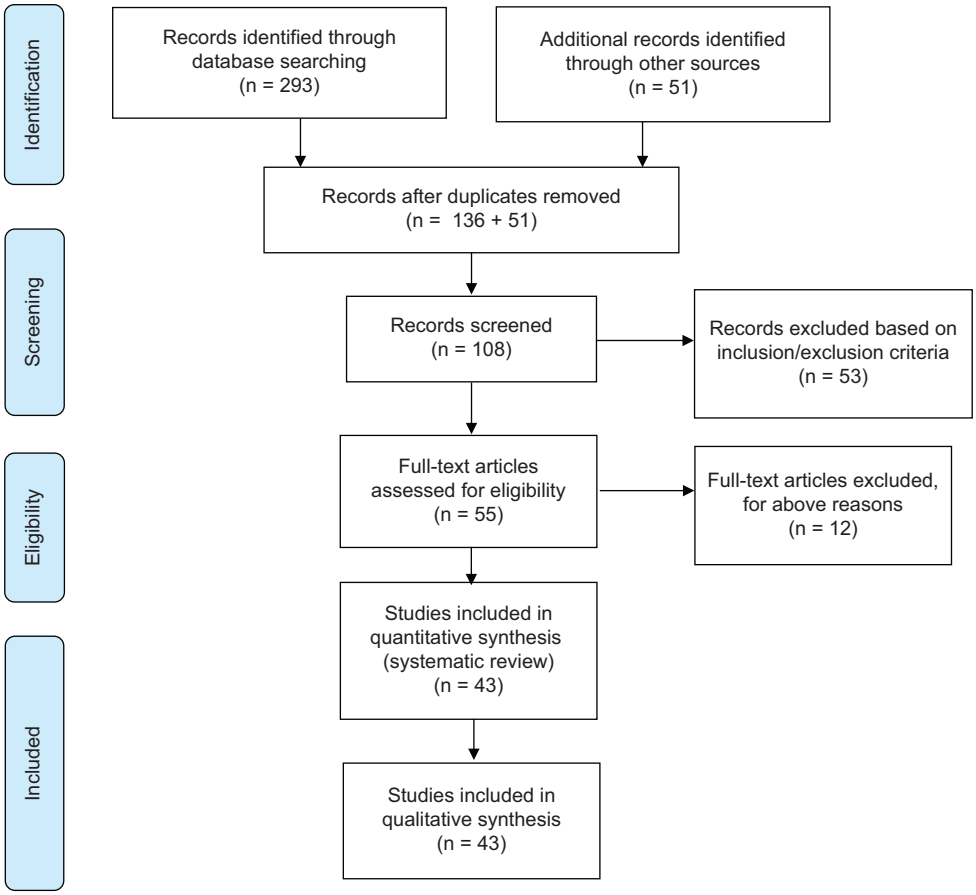
Supplementary Table 1 PRISMA checklist [10]

Section and Topic	Item #	Checklist item	Location where item is reported
		TITLE	
Title	1	Identify the report as a systematic review.	Pg 1
		ABSTRACT	
Abstract	2	See the PRISMA 2020 for Abstracts checklist.	Pg 3
		INTRODUCTION	
Rationale	3	Describe the rationale for the review in the context of existing knowledge.	Pg 4
Objectives	4	Provide an explicit statement of the objective (s) or question (s) the review addresses.	Pg 4
		METHODS	
Eligibility criteria	5	Specify the inclusion and exclusion criteria for the review and how studies were grouped for the syntheses.	Pg 5-7
Information sources	6	Specify all databases, registers, websites, organisations, reference lists and other sources searched or consulted to identify studies. Specify the date when each source was last searched or consulted.	Pg 5-7
Search strategy	7	Present the full search strategies for all databases, registers and websites, including any filters and limits used.	Pg 5-7
Selection process	8	Specify the methods used to decide whether a study met the inclusion criteria of the review, including how many reviewers screened each record and each report retrieved, whether they worked independently, and if applicable, details of automation tools used in the process.	Pg 5-7
Data collection process	9	Specify the methods used to collect data from reports, including how many reviewers collected data from each report, whether they worked independently, any processes for obtaining or confirming data from study investigators, and if applicable, details of automation tools used in the process.	Pg 5-7
Data items	10a	List and define all outcomes for which data were sought. Specify whether all results that were compatible with each outcome domain in each study were sought (e.g. for all measures, time points, analyses), and if not, the methods used to decide which results to collect.	Pg 5-7
	10b	List and define all other variables for which data were sought (e.g. participant and intervention characteristics, funding sources). Describe any assumptions made about any missing or unclear information.	Pg 5-7
Study risk of bias assessment	11	Specify the methods used to assess risk of bias in the included studies, including details of the tool (s) used, how many reviewers assessed each study and whether they worked independently, and if applicable, details of automation tools used in the process.	Pg 5-7
Effect measures	12	Specify for each outcome the effect measure (s) (e.g. risk ratio, mean difference) used in the synthesis or presentation of results.	Pg 5-7
Synthesis methods	13a	Describe the processes used to decide which studies were eligible for each synthesis (e.g. tabulating the study intervention characteristics and comparing against the planned groups for each synthesis (item #5)).	Pg 5-7
	13b	Describe any methods required to prepare the data for presentation or synthesis, such as handling of missing summary statistics, or data conversions.	Pg 5-7
	13c	Describe any methods used to tabulate or visually display results of individual studies and syntheses.	Pg 5-7
	13d	Describe any methods used to synthesize results and provide a rationale for the choice (s). If meta-analysis was performed, describe the model (s), method (s) to identify the presence and extent of statistical heterogeneity, and software package (s) used.	Pg 5-7
	13e	Describe any methods used to explore possible causes of heterogeneity among study results (e.g. subgroup analysis, meta-regression).	Pg 5-7
	13f	Describe any sensitivity analyses conducted to assess robustness of the synthesized results.	Pg 5-7

(*Contd...*)

Supplementary Table 1 (*Continued*)

Section and Topic	Item #	Checklist item	Location where item is reported
		METHODS	
Reporting bias assessment	14	Describe any methods used to assess risk of bias due to missing results in a synthesis (arising from reporting biases).	Pg 5-7
Certainty assessment	15	Describe any methods used to assess certainty (or confidence) in the body of evidence for an outcome.	Pg 5-7
		RESULTS	
Study selection	16a	Describe the results of the search and selection process, from the number of records identified in the search to the number of studies included in the review, ideally using a flow diagram.	Pg 8-9
	16b	Cite studies that might appear to meet the inclusion criteria, but which were excluded, and explain why they were excluded.	Pg 8-9
Study characteristics	17	Cite each included study and present its characteristics.	Pg 8-9
Risk of bias in studies	18	Present assessments of risk of bias for each included study.	Pg 8-9
Results of individual studies	19	For all outcomes, present, for each study: (a) summary statistics for each group (where appropriate) and (b) an effect estimate and its precision (e.g. confidence/credible interval), ideally using structured tables or plots.	Pg 8-9
Results of syntheses	20a	For each synthesis, briefly summarise the characteristics and risk of bias among contributing studies.	Pg 8-9
	20b	Present results of all statistical syntheses conducted. If meta-analysis was done, present for each the summary estimate and its precision (e.g. confidence/credible interval) and measures of statistical heterogeneity. If comparing groups, describe the direction of the effect.	Pg 8-9
	20c	Present results of all investigations of possible causes of heterogeneity among study results.	Pg 8-9
	20d	Present results of all sensitivity analyses conducted to assess the robustness of the synthesized results.	Pg 8-9
Reporting biases	21	Present assessments of risk of bias due to missing results (arising from reporting biases) for each synthesis assessed.	Pg 8-9
Certainty of evidence	22	Present assessments of certainty (or confidence) in the body of evidence for each outcome assessed.	Pg 8-9
		DISCUSSION	
Discussion	23a	Provide a general interpretation of the results in the context of other evidence.	Pg 10-17
	23b	Discuss any limitations of the evidence included in the review.	Pg 17-18
	23c	Discuss any limitations of the review processes used.	Pg 17-18
	23d	Discuss implications of the results for practice, policy, and future research.	Pg 17-18
		OTHER INFORMATION	
Registration and protocol	24a	Provide registration information for the review, including register name and registration number, or state that the review was not registered.	Not registered
	24b	Indicate where the review protocol can be accessed, or state that a protocol was not prepared.	Not prepared
	24c	Describe and explain any amendments to information provided at registration or in the protocol.	
Support	25	Describe sources of financial or non-financial support for the review, and the role of the funders or sponsors in the review.	Pg 1
Competing interests	26	Declare any competing interests of review authors.	Pg 1
Availability of data, code and other materials	27	Report which of the following are publicly available and where they can be found: template data collection forms; data extracted from included studies; data used for all analyses; analytic code; any other materials used in the review.	Template forms and data available on requested



Supplementary Figure 1 PRISMA 2009 flow diagram