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SYSTEMATIC REVIEWS

Genetic studies in irritable bowel syndrome-status quo

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Abstract

AIM

To evaluate the most common studied genetic polymorphisms that may have an etiological role in irritable bowel syndrome (IBS).

METHODS

The data base PubMed was searched for studies analyzing the association between gene polymorphisms and IBS. All original full papers, written in English, were retained for further analysis. The retrieved papers were further systematized according to those polymorphisms that have been detected in IBS.

RESULTS

Considering these criteria, our literature search found 12 polymorphisms, residing in 10 genes, which were reported to be consistently associated with IBS. The initial search identified 189 articles, out of which 48 potentially appropriate articles were reviewed. Of these 48 articles, 41 articles were included in the review. These articles were published between 2002 and 2016. Out of these 41 studies, 17 reported analysis of the serotonin transporter (*SERT*) gene (*SLC6A4*), eight on guanine nucleotide-binding protein subunit beta-3 (*GNbeta3*), six on the serotonin type 3 receptor genes (*HTR3A*), four on (*HTR3E*), three on (*HTR2A*), three the tumor necrosis factor superfamily member TL1A gene (*TNFSF15*), and ten on genetic polymorphisms with limited evidence.

CONCLUSION

Current evidence for the relation between genetic polymorphisms and IBS is limited owing to the fact

that high-quality prospective studies and detailed phenotyping of patients suffering from IBS and matched controls were lacking in the past.

Key words: Irritable bowel syndrome; Gene; Genetic polymorphisms

Core tip: The main genetic polymorphisms encountered in irritable bowel syndrome (IBS) are: Serotonin transporter (*SERT*) gene (*SLC6A4*), guanine nucleotide-binding protein subunit beta-3 (*GNbeta3*), serotonin type 3 receptor genes (*HTR3A*), (*HTR3E*), (*HTR2A*), the tumor necrosis factor superfamily member TL1A gene (*TNFSF15*). We performed a review of existent data, that studied genetic polymorphisms in IBS patients. We found that the actual IBS subgroups are not sufficient in order to identify distinct phenotypes and further in leading to new guiding principles for treatment. This systematic review demonstrates the need for genetic studies with an increasing number of subjects, because contradictory findings in terms of IBS subtype have been reported.

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INTRODUCTION

Irritable bowel syndrome (IBS) is the main digestive functional disorder, with a prevalence of 10%-20% of the population and has multifactorial etiology since genetic predisposition and environmental factors shape the phenotype.

According to the Rome Ⅳ criteria, the syndrome is defined as recurrent abdominal pain on average at least 1 d/wk in the last 3 mo, associated with two or more of the following symptoms: related to defecation, associated with a change in the frequency of stool, associated with a change in form (consistency) of stool. Classifying patients with IBS into specific subtypes based on predominant bowel habits is useful because is focusing the treatment on the predominant symptom. Accordingly to the Rome Ⅳ Criteria, IBS is classified into four subtypes: IBS with predominant constipation (IBS-C), IBS with predominant diarrhea (IBS-D), with mixed bowel habits (IBS-M) or unsubtyped (IBS-U). Patients meet diagnostic criteria for IBS-U if their bowel habits cannot be accurately categorized in any of the above subtypes[1]. The genetic predisposition is underlying the pathogenesis and the pathophysiology of IBS. Studies that point out higher concordance rates of monozygotic twins compared to dizygotic twins suggest that there may be distinct molecular bases for all IBS subtypes and genes that control neuronal function, the epithelial barrier integrity, mucosal immune interactions with bacteria in the gut. Unfortunately, the number of studies about single nucleotide polymorphisms (SNP) in selected candidate genes associated with IBS is still small.

The aim of this study was to review the existing literature on genetic polymorphisms associated with IBS.

MATERIALS AND METHODS

A PubMed search was carried out in September 2016, looking for published papers analyzing the association between gene polymorphisms and IBS. Search keywords were: *IBS* and gene *polymorphism.* The inclusion criteria were: original articles that included patients with IBS-C, IBS-D or IBS-M, and that studied genetic polymorphisms in IBS patients. Exclusion criteria were: reviews, lack of abstract, non-English publications. Furthermore, ethical background was taken into account. We decided not to analyze SNP, which is less investigated, we only found it reported in five papers, because we decided that they are not relevant and may introduce bias.

RESULTS

As a result of our literature survey, we were able to review 12 polymorphisms, residing in 10 genes. All of them are considered to be associated with IBS (Table 1). The initial search identified 182 articles, out of which 48 potentially appropriate articles were reviewed. Of these 48 articles, 44 articles were included in the review. These articles were published between 2002 and 2016. Out of these 44 studies, 20 reported analysis of the serotonin transporter (*SERT*) gene (*SLC6A4*), eight on guanine nucleotide-binding protein subunit beta-3 (*GNbeta3*), six on the serotonin type 3 receptor gene (*HTR3A*), four on (*HTR3E*), three on (*HTR2A*), three the tumor necrosis factor superfamily member TL1A gene (*TNFSF15*), and ten on genetic polymorphisms with limited evidence (Figure 1).

In the following we will describe the reported evidence of a relationship between gene polymorphisms and IBS published to date.

Serotonin transporter gene

Serotonin (5-hydroxtryptamine, *5-HT*) is an essential neurotransmitter involved in regulation of gut function, by playing key roles in intestinal peristalsis and in sensory functions mediated *via* the brain-gut axis. The serotonin transporter (*SERT*) encoded by the gene *SLC6A4* regulates the intensity and duration of serotonin signaling by reuptaking serotonin from the synaptic cleft, thereby terminating its efficacy. This makes it an excellent candidate gene for analysis of genetic predisposition to IBS.

Disturbance in serotonin reuptake can modify enteric signaling, leading to gut dysfunctions, thereby

Table 1 Number of articles, analyzing the relation between genetic polymorphisms and irritable bowel syndrome

Gene	SNP	Polymorphism	IBS type	Diagnostic criteria	Number of articles	Ref.
SLC6A4	rs4795541	5-HTTLPR (-1950- 1949insT, -1950-1949insC), STin2.9 VNTR	IBS-C	Rome Ⅰ, Ⅱ, Ⅲ	15	[2-5,7-12,14,15,19,22,27]
	rs25531	179A > G (-1936A > G)		Rome Ⅱ, Ⅲ	4	[8-10,17]
HTR2A	rs6311	-1438G > A (-998G > A	IBS-D	Rome Ⅰ, Ⅱ, Ⅲ	1	[4]
	rs6313	102C > T	IBS-D	Rome Ⅰ, Ⅱ, Ⅲ	2	[4,35]
HTR3A	rs1062613	42C > T; 178C > T (-24C > T)	IBS-D	Rome Ⅰ, Ⅱ, Ⅲ	6	[2,16,20-22,45]
HTR3E	rs56109847	76G > A	IBS-D	Rome Ⅰ, Ⅱ, Ⅲ	5	[16,18,20,21,45]
GNB3	rs5443	825C > T	IBS-C	Rome Ⅱ, Ⅲ	8	[23-25,29,32,34,35]
TNFSF15	rs4263839	A/G	IBS-C	Rome Ⅰ, Ⅱ, Ⅲ	3	[6,26,28]
Limited number of studies: *pV158M CCK rec.intron1 NXPH1CDC42*				Rome Ⅲ	10	[30,31,33,36-40,43,44]

IBS: Irritable bowel syndrome; SNP: Single nucleotide polymorphisms.

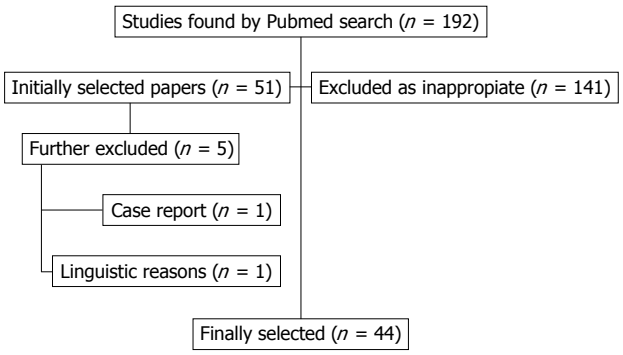

Figure 1 Results of PubMed search and selection of original articles included in the review.

contributing to the pathophysiology of IBS.

The solute carrier family 6 member 4 (*SLC6A4*) gene encodes the serotonin transporter (*SERT*). Polymorphisms in the promoter region of the SERT gene has a direct effect on transcriptional activity, which may result in altered 5-HT reuptake activity. The investigation of the association between *5HTTLPR* in the *SERT* gene and IBS, using subgroup population-based analysis, point out that visceral hypersensitivity in IBS can be related to genetic factors[2-6]. To date, the S allele in the promoter region as well as the *STin2.9 VNTR* allele residing in an intron, have been reported to be related to anxiety and depression, a result that supports a biopsychosocial model of IBS, with the genotype in *SLC6A4* that is increasing the risk for depressive episodes. Increased risk of IBS-C is presented by individuals with of L/L genotype and 12/12-L/L genotype association[5,6]. IBS-D and IBS-A are more frequent in individuals with L/S genotype[6]. Other studies suggest that the s/L polymorphism of serotonin transporter gene is linked only with the IBS-C development, this link being present only in East Asian population[7]. Moreover, the response to tegaserod was influenced by the genotype: L/L being poorer than S/S and S/L genotypes[5]. Carriers of S allele in *5-HTTLPR* region was published as being frequent in Chinese Han population, with IBS, but other associations studies looking for IBS and variable number of tandem repeats (*VNTRs*) and tag SNPs, such as rs1042173, rs3794808, rs2020936 in *SERT* gene [using polymerase chain reaction (PCR) and TaqMan® SNP Genotyping, and positive haplotype], were not found[8]. *SLC6A4*-polymorphism and higher levels of 5-HT (in rectal biopsy of patients) were significantly linked with IBS-D and abdominal pain, suggesting that *SLC6A4* has an important role in IBS pathophysiology[9,10]. Also in IBS-D, platelet *SERT* is reduced and is related with low levels of *SERT* mRNA.

A metaanalysis by Zhang *et al*[11] looked to 25 studies including more than 3000 patients with IBS and more than 3000 controls (diagnosed with different criteria according to the moment of the study: Rome Ⅰ/Ⅱ/Ⅲ). The meta-analysis showed that the *5HTTLPR L* allele and L/L are involved in the IBS-C development, in East Asian population, but not Central Asian populations.

On the contrary, other studies found a negative association between IBS and 5HTTLPR in the SERT gene. This is the case of a metaanalysis by Areeshi *et al*[12], which analyzed 12 studies with over 2000 IBS cases and over 2000 The same lack of association is found in the studies that have examined another *SERT* gene polymorphism, STin2 (located in intron 2), with undetermined ethnicity[13].

A study on a group of North American Caucasian female patients with IBS-D, analyzed leukocyte DNA, by polymerase chain reaction, for nine *SERT* polymorphisms. The result was that *SERT-P* S/S genotype was significant associated with IBS-D[14]. On the other hand, a study on American and Asian populations demonstrates that *SLC6A4* (S/L) polymorphism is associated with reduced risk of IBS[15].

The activation of different brain regions during colorectal distension in subjects carrying the S allele of the *SERT* gene *SLC6A4* promoter polymorphism 5-HTTLPR, suggests that individuals with a reduced level of *SERT* may more intensively respond to gut signals in emotion-regulating brain circuit. The amygdala region is more activated during a fearful face recognition paradigm in fMRI studies. This data demonstrates the relation between visceral pain and the individuals with a weak function of serotonin transporter[23]. A study using the Rome Ⅰ criteria, in 54 Turkish IBS patients, showed

a high incidence of the C/C genotype for 102T > C, A/A genotype for−1438G > A, *HTR2A* gene, rs6313 and IBS-D[3]. Similar results were found in a Greek study, showing that the frequencies of the SS genotype and S allele of the serotonin transporter polymorphism were significantly associated with IBS and the TT genotype and T allele frequencies of G protein β3 subunit showed also significant difference between the IBS patients and healthy controls[2].

Other plausible candidates of the serotonergic system represent $5HT_3$ receptors ($5HT_3Rs$) mediating the effects of 5HT on intestinal functions during the postprandial period. A sequencing study of the *HTR3* genes in IBS detected the 5'-UTR variant c.-42C > T of *HTR3A* (rs1062613) and 3'-UTR variant c. 76G > A in *HTR3E* (rs62625044). They found an association of SNPs in *HTR3A* and *HTR3E* in patients with IBS-D in a cohort from the United Kingdom; in particular the SNP in *HTR3E* was replicated in another cohort from Germany[2,16]. A recent study that investigated the relation between these SNPs in *HTR3A* and *HTR3E* and IBS-D in 500 IBS-D Chinese patients and 500 healthy control subjects replicated these findings. The PCR-RFLP method revealed a significant difference in the SNP frequency between the IBS-D patients and the healthy control subjects in the distribution of genotype and the minor allele of rs1062613 in *HTR3A* gene. Moreover, data about rs62625044 in *HTR3E* gene, evidenced a significant difference between the distribution of GA genotype and A allele, only in female patients[16].

A small sample size study of pacients with IBS showed that the carriers of the rare G allele of rs25531 had approximately threefold increased odds to present IBS than healthy controls . Onwards, the G-allele was more frequent in diarrhea-predominant subjects than in constipation-predominant or alternator subjects[17].

Recent studies demonstrated that a functional variant (rs56109847) in the 3'-untranslated regions (3'-UTR) of the serotonin receptor 3E (HTR3E) gene associated with IBS-D in British populations is also present in IBS-D in the Chinese females, emphasizing the role of miR-510 on $5\text{-}HT_{3E}$ expression of colonic tissues in patients with gastrointestinal disorders. Moreover, the mechanism that underlies the association of HTR3E SNP rs56109847 with IBS-D is also described. The $5\text{-}HT_{3E}$ rs56109847 could directly inhibit the binding of miR-510 to HTR3E 3'-UTR in HEK293 and HT-29 cells and confirmed that the SNP (rs56109847) of the non-coding region of HTR3E affected the binding of mircoRNA, thus affecting the permeability of the GI tract[18].

In contradiction with the analysed data, a study shows that there is no association between the genetic polymorphism in the SERT-P gene and IBS. The fact that SERT-P polymorphism has recently been associated with treatment response is a further proof that the genetic polymorphism in the SERT-P gene might have a pharmacogenetic role[19].

Another more recent study replicated these findings in patients with IBS-D from Yangzhou, Jiangsu province, showing a significant difference between patients and the controls in *HTR3A* (rs1062613) and the frequency of T allele was significantly higher in both female and male patients than that in the controls ($P < 0.05$). They performed polymerase chain reaction (PCR) amplification and restriction fragment length polymorphism (RFLP) technique on DNAs from 300 healthy subjects and 450 patients with IBS-D[20]. Of note, the SNPs rs1062613 in *HTR3A* has initially been associated with major depression and "harm avoidance", an inherited trait associated with depression and anxiety, frequently encountered in IBS. In a study from 2011, this SNP has been correlated with the severity of IBS symptoms, anxiety and changes in amygdala activity[15]. Alosetron, a selective $5HT_3R$ antagonist, beneficial in the management of symptoms like abdominal cramping, stool urgency and diarrhea in women with IBS-D was investigated in a pharmacogenetics study[21]. This revealed a greater efficacy of slowing down colonic transit as evidenced by the fact that L/L compared to L/S or S/S carriers benefitted from the treatment, by being high responders. This seems to be plausible based on the hypothesis that L/L carriers, who are supposed to present with increased SERT expression, and consequently 5HT reuptake, may present lower synaptic 5HT levels and therefore less competition between endogenous 5HT and alosetron[22].

Catechol-O-methyltransferase (COMT) is an enzyme that degrades dopamine, epinephrine, norepinephrine and the functional polymorphism pV158M has most extensively analyzed to date in various conditions. The Val alleles lead to four-fold higher enzymatic activity compared to the Met allele and thereby may influence metabolic levels of its substrates[23]. The gene variant has been demonstrated, to play an essential role in processes associated with abstract thought, task structure, and the placebo effect[23,24].

It is well established that depression, anxiety and pain syndromes are related to altered COMT activity, conditions showing also a high co-morbidity with IBS. Consequently it presented another plausible candidate to be explored in the context of IBS. In a recent study from Sweden, the V/V genotype had a significantly higher occurrence compared with controls, but V/M genotype, had a lower occurrence in IBS compared with controls and exhibited significantly increased bowel frequency[24]. In elderly Chinese patients (over the age of 60 years), *COMT158Met* was related with IBS and significantly more prevalent in patients with IBS-D. Furthermore, it was prevalent in those patients with symptomatology that persisted over 5 years[25].

Tumor necrosis factor superfamily-15

Tumor necrosis factor superfamily-15 gene (*TNFSF15*, also known as *VEGI* or *TL1A*) is a cytokine that has main functions in angiogenesis, immune system mobilization and inflammation. *TNFSF15* stimulates T cell activation, Th1 cytokine production, dendritic cell maturation and inhibits endothelial cell proliferation and endothelial

progenitor cell differentiation. The risk allele of the SNP rs4263839 G in *TNFSF15* was initially associated with an increased risk of IBS, more pronouncedly, IBS-C[6].

In respect to postinfectious IBS(PI-IBS) it has been hypothesized that polymorphisms in genes whose expression were altered by gastroenteritis might be linked to IBS with diarrhea (IBS-D) which closely resembles PI-IBS[25]. Han *et al*[25] established an IBS-D association with rs6478109 and rs6478108, which are in linkage disequilibrium with rs4263839. In fact, they found indeed IBS-D and PI-IBS patients to be associated with *TNFSF15* and TNF α genetic polymorphisms which also predispose to Crohn's disease suggesting a possible common underlying pathogenesis. In addition, both SNPs are associated with TNFSF15 expression in colorectal tissue[27]. Furthermore, Czogalla *et al*[2] recently confirmed a modest association (OR 1.24) in IBS-C in a meta-analysis combining own validation data with published data from the two previous studies.

TL1A-Death Receptor 3 has an essential role in production of interferon-c and interleukin-17 *via* proliferation and differentiation of T-helper 17, explaining patterns of immune response in host-microbiota interaction with commensal bacteria that contribute to IBS risk. As well, data shows its implication in other inflammatory disorders[26-28].

Guanine nucleotide-binding protein

Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-3 (GNβ3) is a protein that is encoded by the gene *GNB3*[28]. The G-protein is an important factor in intracellular signal transduction, mediating functions of ion channels and protein kinases. The *SNP*-825C > T is leading to a modified signal transduction of functional impact: changes of sensory function or motility associated with FGID (functional gastrointestinal disorders)[29]. The association of this polymorphism with IBS has been demonstrated, and recent data shows that alteration of *GNB3* 825C > T CC type has a direct effect on gastrointestinal sensitivity and peristalsis[30]. A group of elderly Chinese IBS evaluated using the Geriatric Depression Scale, was not able to relate the *GNB3*-825C > T *SNP* with IBS[25,31]. However, the TC/TT genotypes are associated with lower sensations of gas and urgency in response to rectal distention after administration of clonidine[32]. In line with this, a study on a group from Korea evidenced that the *GNB3* 825C > TT allele is associated with IBS-C and studies analyzing patients from Greece, also confirmed that the TT genotype and *GNB3* T allele have a significant association with IBS[32-34].

A study in which two large independent IBS cohorts were genotyped to assess genetic variability in immune, neuronal and barrier integrity genes, determined that the following SNPs associated independently: rs17837965-*CDC42* with IBS-C (OR exploratory = 1.59 (1.05 to 1.76); OR validation = 1.76 (1.03 to 3.01)) and rs2349775-*NXPH1* with IBS-D (OR exploratory = 1.28 (1.06 to 1.56); OR validation = 1.42 (1.08 to 1.88)). The study included 935 IBS patients, 639 controls and 384 single nucleotide polymorphisms (SNPs) covering 270 genes. Other three SNPs in immune-related genes (rs1464510-*LPP*, rs1881457-*IL13*, rs2104286-*IL2RA*), one SNP in a neuronal gene (rs2349775-*NXPH1*) and two SNPs in epithelial genes (rs245051-*SLC26A2*, rs17837965-*CDC42*) were weakly associated with IBS ($P < 0.05$)[34-45].

DISCUSSION

The present review identified articles, most of them prospective studies, on genetic polymorphisms in IBS pathogenesis or after therapy. The major pitfall is that patients were recruited based on a non-uniform symptom classification: Rome Ⅰ, Rome Ⅱ, or Rome Ⅲ in the studies that were taken into account. Study limitations were represented by language barriers of some articles (which prevented access), and the low number of patients involved in most of the studies (underpowered); the main reason for excluding articles was the insufficient number of studies on a particular genetic polymorphism and articles written in non-English publications. Other limitations of the meta-analysis were the intricacy of ethnicities, and the difficulty of taking multiple genotypes testing into account. Above all that, statistical results were rarely corrected for multiplicity. As a result false positive associations may have been reported.

The polymorphisms of the Serotonin transporter (SERT or SLC6A4) gene are the most frequent genetic polymorphisms studied in IBS to date. Studies proved that the A allele of *HTR3E* was significantly higher in female IBS-D patients and there were no differences in either A allele or GA genotype between male patients. A possible reason for why there is no association to be found in male, can be explained by the effect of ovarian hormones on visceral sensitivity. A supposition which needs to be verified by future research.

A recent meta-analysis of immunogenetic case–control association studies in IBS confirmed a moderate association of rs4263839 in *TNFSF15*, and particularly with IBS-C. Control samples recruited by harmonized criteria are essential in order to overcome limitations like low statistical power and large heterogeneity for studies of IBS.

Because of the limited number of studies, further studies are needed for the following polymorphisms: Cholecystokinin (CCK) is a peptide hormone responsible for stimulating the digestion of fat and protein and is produced by I-cells in the mucosal epithelium of the small bowel. It has the effect of releasing digestive enzymes and bile from the pancreas and gallbladder and recent data evidenced that low densities of secretin and CCK cells in IBS-diarrhea patients can cause a functional pancreatic insufficiency and also inadequate gall emptying[27,28].

Polymorphism in CCK receptor intron 1 was associated with IBS-C and IBS-M in Korean population[37,38]. Also with limited evidence is the adhesion between dendrites

and axons, that is promoted by a tight complex with alpha neurexins and neurexophilin-1 a protein encoded by the *NXPH1* gene. Genetic variants in *NXPH1* are associated with IBS-D[6]. Cell division control protein 42 homolog (CDC42) is a protein with an essential role in cell cycle regulation, including cell structure, migration, endocytosis and cell cycle progression. Genetic variants in CDC42 are associated with IBS-D[39].

The biopsychosocial model of illness and disease, as first described by Engel, reconcileed the dualistic concept that separated illness and disease and is a good way to explain the interaction between cultural factors, ethnicity, geographic region, types of food, endocrinological factors, immunological factors and genetic markers, which exist in patients with IBS. Recent studies analyzing individual coping strategies, cultural level, education level, religious beliefs about health and disease, demonstrated that a biopsychosocial conceptualization of the pathogenesis and clinical expression of IBS is mandatory. Further, somatic symptoms interact with the psychological status and promote each other, making the investigation of IBS more difficult[40].

A recent study that analysed 288 103 participants from 41 countries, showed that the global prevalence of IBS has a significant degree of heterogeneity that ranged from 1.1% in France and Iran to 35.5% in Mexico, with significant variance in regional prevalence rates, from 17.5% (95%CI: 16.9% to 18.2%) in Latin America, 9.6% (9.5% to 9.8%) in Asia, 7.1% (8.0% to 8.3%) in North America/Europe/Australia/New Zealand, to 5.8% (5.6% to 6.0%) in the Middle East and Africa[41].

A major pitfall in the current genetic studies in IBS is represented by the low number of subjects included in the majority of studies. Fortunately, the number of centers around the world that are collecting samples is growing. Nevertheless no unified genetics workflow existed. From the genetic perspective, the actual IBS subgroups are not sufficient in order to identify distinct phenotypes and further in leading to new guiding principles for treatment. These limitations can be overcome by international cooperation, like the GENIEUR network (Genes in Irritable Bowel Syndrome Research Network Europe, www.GENIEUR.eu), who allows the contribution of specialists from many countries and the collecting of large samples of subjects[42] who are deeply phenotyped to allow genotype phenotype correlation and data mining approached[42]. Such studies allow also the standardization of investigative tools in the approach of IBS patients[43-45].

In conclusion, Current evidence for the relation between genetic polymorphisms and IBS is limited owing to the fact that high-quality prospective studies and detailed phenotyping of patients suffering from IBS and matched controls were lacking in the past. Studies on functional gastrointestinal disorders and genetic polymorphisms analyzing the same genetic variants in comparably characterized case control cohorts are also very limited. Furthermore, association of *TNFSF15* genetic polymorphisms, which also predispose to Crohn's disease, suggest a possible common underlying pathogenesis. However, for both polymorphisms contradictory findings in terms of IBS subtype have been reported underlining the necessity of more detailed phenotypic information for data stratification. To date, the s/l polymorphism in *SLC6A4,* represents the most frequently studied polymorphism and the *HTR3E* SNP has been replicated in four studies to date.

ARTICLE HIGHLIGHTS

Research background

The irritable bowel syndrome (IBS) is a hot topic and the uncovering its genetic determination is very important.

Research motivation

Knowing the genetic link in the occurrence of IBS could offer the perspective to better know this condition and to improve its management.

Research objectives

In order to shed light on this topic, we carried out a systematic review of the data on main genetic polymorphisms described uptoday.

Research methods

A PubMed search was carried out in September 2016, looking for studies analyzing the association between gene polymorphisms and IBS. Search keywords were: IBS and gene polymorphism. The inclusion criteria were: original articles that included patients with IBS-C, IBS-D or IBS-M, and that studied genetic polymorphisms in IBS patients. Exclusion criteria were: reviews, lack of abstract, non-English publications.

Research results

The result of our study was a review of 12 polymorphisms, residing in 10 genes reported to be associated with the pathogenesis and the pathophysiology of IBS. The main problem that remains to be solved in the current genetic studies analysing IBS is represented by the low number of subjects included in the majority of studies.

Research conclusions

High-quality evidence for the relation between genetic polymorphisms and the IBS etiology is lacking, as a result of the insufficient number of high-quality prospective studies. Similar studies on functional gastrointestinal disorders and genetic polymorphisms are also very limited. The strength of articles, included in this review are the determination of each genetic polymorphism, using high efficiency techniques. The polymorphisms of the Serotonin transporter (*SERT* or *SLC6A4*) gene were the most frequent genetic polymorphisms studied in this pathology. Investigation of PI-IBS patients showed associations with *TNFSF15* genetic polymorphisms which also predispose to Crohn's disease suggesting a possible common underlying pathogenesis.

Research perspectives

From the genetic perspective, the actual IBS subgroups are not sufficient in order to identify distinct phenotypes and further in leading to new guiding principles for treatment. These limitations can be overcome by international cooperation, like the GENIEUR network (Genes in Irritable Bowel Syndrome Research Network Europe), who allows the contribution of specialists from many countries and the collecting of large samples of subjects who are deeply phenoytped to allow genotype phenotype correlation and data mining approached. Such studies allow also the standardization of investigative tools in the approach of IBS patients.

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REFERENCES

1 **Schmulson MJ**, Drossman DA. What Is New in Rome IV. *J Neurogastroenterol Motil* 2017; **23**: 151-163 [PMID: 28274109 DOI: 10.5056/jnm16214]

2 **Czogalla B**, Schmitteckert S, Houghton LA, Sayuk GS, Camilleri M, Olivo-Diaz A, Spiller R, Wouters MM, Boeckxstaens G, Bermejo JL, Niesler B. A meta-analysis of immunogenetic Case-Control Association Studies in irritable bowel syndrome. *Neurogastroenterol Motil* 2015; **27**: 717-727 [PMID: 25824902 DOI: 10.1111/nmo.12548]

3 **Fukudo S**, Kanazawa M, Mizuno T, Hamaguchi T, Kano M, Watanabe S, Sagami Y, Shoji T, Endo Y, Hongo M, Itoyama Y, Yanai K, Tashiro M, Aoki M. Impact of serotonin transporter gene polymorphism on brain activation by colorectal distention. *Neuroimage* 2009; **47**: 946-951 [PMID: 19426812 DOI: 10.1371/journal.pone.0123183]

4 **Pata C**, Erdal E, Yazc K, Camdeviren H, Ozkaya M, Ulu O. Association of the -1438 G/A and 102 T/C polymorphism of the 5-Ht2A receptor gene with irritable bowel syndrome 5-Ht2A gene polymorphism in irritable bowel syndrome. *J Clin Gastroenterol* 2004; **38**: 561-566 [PMID: 15232358 DOI: 10.1097/00004836-200408000-00005]

5 **Li YY**, Nie YQ, Xie J, Tan HZ, Zhou YJ, Wang H. Serotonin transporter gene polymorphisms in irritable bowel syndrome and their impact on tegaserod treatment. *Zhonghua Neike Zazhi* 2006; **45**: 552-555 [PMID: 17074108]

6 **Zucchelli M**, Camilleri M, Andreasson AN, Bresso F, Dlugosz A, Halfvarson J, Törkvist L, Schmidt PT, Karling P, Ohlsson B, Duerr RH, Simren M, Lindberg G, Agreus L, Carlson P, Zinsmeister AR, D'Amato M. Association of TNFSF15 polymorphism with irritable bowel syndrome. *Gut* 2011; **60**: 1671-1677 [PMID: 21636646 DOI: 10.1136/gut.2011.241877]

7 **Saito YA**. Genes and irritable bowel syndrome: is there a link? *Curr Gastroenterol Rep* 2008; **10**: 355-362 [PMID: 18627646 DOI: 10.1007/s11894-008-0069-9]

8 **Yuan J**, Kang C, Wang M, Wang Q, Li P, Liu H, Hou Y, Su P, Yang F, Wei Y, Yang J. Association study of serotonin transporter SLC6A4 gene with Chinese Han irritable bowel syndrome. *PLoS One* 2014; **9**: e84414 [PMID: 24392134 DOI: 10.1371/journal.pone.0084414]

9 **Kumar S**, Ranjan P, Mittal B, Ghoshal UC. Serotonin transporter gene (SLC6A4) polymorphism in patients with irritable bowel syndrome and healthy controls. *J Gastrointestin Liver Dis* 2012; **21**: 31-38 [PMID: 22457857]

10 **Foley S**, Garsed K, Singh G, Duroudier NP, Swan C, Hall IP, Zaitoun A, Bennett A, Marsden C, Holmes G, Walls A, Spiller RC. Impaired uptake of serotonin by platelets from patients with irritable bowel syndrome correlates with duodenal immune activation. *Gastroenterology* 2011; **140**: 1434-1443.e1 [PMID: 21315720 DOI: 10.1053/j.gastro.2011.01.052]

11 **Zhang ZF**, Duan ZJ, Wang LX, Yang D, Zhao G, Zhang L. The serotonin transporter gene polymorphism (5-HTTLPR) and irritable bowel syndrome: a meta-analysis of 25 studies. *BMC Gastroenterol* 2014; **14**: 23 [PMID: 24512255 DOI: 10.1186/1471-230X-14-23]

12 **Areeshi MY**, Haque S, Panda AK, Mandal RK. A serotonin transporter gene (SLC6A4) polymorphism is associated with reduced risk of irritable bowel syndrome in American and Asian population: a meta-analysis. *PLoS One* 2013; **8**: e75567 [PMID: 24069428 DOI: 10.1371/journal.pone.0075567]

13 **Pata C**, Erdal ME, Derici E, Yazar A, Kanik A, Ulu O. Serotonin transporter gene polymorphism in irritable bowel syndrome. *Am J Gastroenterol* 2002; **97**: 1780-1784 [PMID: 12135035 DOI: 10.1111/j.1572-0241.2002.05841.x]

14 **Yeo A**, Boyd P, Lumsden S, Saunders T, Handley A, Stubbins M, Knaggs A, Asquith S, Taylor I, Bahari B, Crocker N, Rallan R, Varsani S, Montgomery D, Alpers DH, Dukes GE, Purvis I, Hicks GA. Association between a functional polymorphism in the serotonin transporter gene and diarrhoea predominant irritable bowel syndrome in women. *Gut* 2004; **53**: 1452-1458 [PMID: 15361494 DOI: 10.1136/gut.2003.035451]

15 **Choi YJ**, Hwang SW, Kim N, Park JH, Oh JC, Lee DH. Association Between SLC6A4 Serotonin Transporter Gene Lainked Polymorphic Region and ADRA2A -1291C> G and Irritable Bowel Syndrome in Korea. *J Neurogastroenterol Motil* 2014; **20**: 388-399 [PMID: 24917480 DOI: 10.5056/jnm14020]

16 **Gu QY**, Zhang J, Feng YC, Dai GR, Du WP. Association of genetic polymorphisms in HTR3A and HTR3E with diarrhea predominant irritable bowel syndrome. *Int J Clin Exp Med* 2015; **8**: 4581-4585 [PMID: 26064388]

17 **Kohen R**, Jarrett ME, Cain KC, Jun SE, Navaja GP, Symonds S, Heitkemper MM. The serotonin transporter polymorphism rs25531 is associated with irritable bowel syndrome. *Dig Dis Sci* 2009; **54**: 2663-2670 [PMID: 19125330 DOI: 10.1007/s10620-008-0666-3]

18 **Zhang Y**, Li Y, Hao Z, Li X, Bo P, Gong W. Association of the Serotonin Receptor 3E Gene as a Functional Variant in the MicroRNA-510 Target Site with Diarrhea Predominant Irritable Bowel Syndrome in Chinese Women. *J Neurogastroenterol Motil* 2016; **22**: 272-281 [PMID: 26787495 DOI: 10.5056/jnm15138]

19 **Van Kerkhoven LA**, Laheij RJ, Jansen JB. Meta-analysis: a functional polymorphism in the gene encoding for activity of the serotonin transporter protein is not associated with the irritable bowel syndrome. *Aliment Pharmacol Ther* 2007; **26**: 979-986 [PMID: 17877505 DOI: 10.1111/j.1365-2036.2007.03453.x]

20 **Zhang Y**, Huang Y, Bo P. Association between diarrhea-predominant irritable bowel syndrome and HTR3A, HTR3E gene polymorphism in Yangzhou, Jiangsu province, China. *Zhonghua Liuxingbingxue Zazhi* 2013; **34**: 721-724 [PMID: 24257177 DOI: 10.3760/cma.j.issn.0254-6450.2013.07.014]

21 **Bleser S**. Alosetron for severe diarrhea-predominant irritable bowel syndrome: improving patient outcomes. *Curr Med Res Opin* 2011; **27**: 503-512 [PMID: 21208139 DOI: 10.1185/03007995.2010.547933]

22 **Camilleri M**, Atanasova E, Carlson PJ, Ahmad U, Kim HJ, Viramontes BE, McKinzie S, Urrutia R. Serotonin-transporter polymorphism pharmacogenetics in diarrhea-predominant irritable bowel syndrome. *Gastroenterology* 2002; **123**: 425-432 [PMID: 12145795 DOI: 10.1053/gast.2002.34780]

23 **Bruder GE**, Keilp JG, Xu H, Shikhman M, Schori E, Gorman JM, Gilliam TC. Catechol-O-methyltransferase (COMT) genotypes and working memory: associations with differing cognitive operations. *Biol Psychiatry* 2005; **58**: 901-907 [PMID: 16043133 DOI: 10.1016/j.biopsych.2005.05.010]

24 **Karling P**, Danielsson Å, Wikgren M, Söderström I, Del-Favero J, Adolfsson R, Norrback KF. The relationship between the val158met catechol-O-methyltransferase (COMT) polymorphism and irritable bowel syndrome. *PLoS One* 2011; **6**: e18035 [PMID: 21437260 DOI: 10.1371/journal.pone.0018035]

25 **Han CJ**, Kohen R, Jun S, Jarrett ME, Cain KC, Burr R, Heitkemper MM. COMT Val158Met Polymorphism and Symptom Improvement Following a Cognitively Focused Intervention for Irritable Bowel Syndrome. *Nurs Res* 2017; **66**: 75-84 [PMID: 28252569 DOI: 10.1097/NNR.0000000000000199]

26 **Ek WE**, Reznichenko A, Ripke S, Niesler B, Zucchelli M, Rivera NV, Schmidt PT, Pedersen NL, Magnusson P, Talley NJ, Holliday EG, Houghton L, Gazouli M, Karamanolis G, Rappold G, Burwinkel B, Surowy H, Rafter J, Assadi G, Li L, Papadaki E, Gambaccini D, Marchi S, Colucci R, Blandizzi C, Barbaro R, Karling P, Walter S, Ohlsson B, Tornblom H, Bresso F, Andreasson A, Dlugosz A, Simren M, Agreus L, Lindberg G, Boeckxstaens

G, Bellini M, Stanghellini V, Barbara G, Daly MJ, Camilleri M, Wouters MM, D'Amato M. Exploring the genetics of irritable bowel syndrome: a GWA study in the general population and replication in multinational case-control cohorts. *Gut* 2015; **64**: 1774-1782 [PMID: 25248455 DOI: 10.1136/gutjnl-2014-307997]

27 **Swan C**, Duroudier NP, Campbell E, Zaitoun A, Hastings M, Dukes GE, Cox J, Kelly FM, Wilde J, Lennon MG, Neal KR, Whorwell PJ, Hall IP, Spiller RC. Identifying and testing candidate genetic polymorphisms in the irritable bowel syndrome (IBS): association with TNFSF15 and TNFα. *Gut* 2013; **62**: 985-994 [PMID: 22684480 DOI: 10.1136/gutjnl-2011-301213]

28 **Weinstein LS**, Chen M, Xie T, Liu J. Genetic diseases associated with heterotrimeric G proteins. *Trends Pharmacol Sci* 2006; **27**: 260-266 [PMID: 16600389 DOI: 10.1016/j.tips.2006.03.005]

29 **Holtmann G**, Gschossmann J, Neufang-Hüber J, Gerken G, Talley NJ. Differences in gastric mechanosensory function after repeated ramp distensions in non-consulters with dyspepsia and healthy controls. *Gut* 2000; **47**: 332-336 [PMID: 10940267 DOI: 10.1136/gut.47.3.332]

30 **Holtmann G**, Talley NJ. Hypothesis driven research and molecular mechanisms in functional dyspepsia: the beginning of a beautiful friendship in research and practice? *Am J Gastroenterol* 2006; **101**: 593-595 [PMID: 16542295 DOI: 10.1111/j.1572-0241.2006.00480.x]

31 **Wang Y**, Wu Z, Qiao H, Zhang Y. A genetic association study of single nucleotide polymorphisms in GNβ3 and COMT in elderly patients with irritable bowel syndrome. *Med Sci Monit* 2014; **20**: 1246-1254 [PMID: 25037115 DOI: 10.12659/MSM.890315]

32 **Camilleri M**, Busciglio I, Carlson P, McKinzie S, Burton D, Baxter K, Ryks M, Zinsmeister AR. Pharmacogenetics of low dose clonidine in irritable bowel syndrome. *Neurogastroenterol Motil* 2009; **21**: 399-410 [PMID: 19309415 DOI: 10.1111/j.1365-2982.2009.01263.x]

33 **Lee HJ**, Lee SY, Choi JE, Kim JH, Sung IK, Park HS, Jin CJ. G protein beta3 subunit, interleukin-10, and tumor necrosis factor-alpha gene polymorphisms in Koreans with irritable bowel syndrome. *Neurogastroenterol Motil* 2010; **22**: 758-763 [PMID: 20337945 DOI: 10.1111/j.1365-2982.2010.01496.x]

34 **Markoutsaki T**, Karantanos T, Gazouli M, Anagnou NP, Ladas SD, Karamanolis DG. Serotonin transporter and G protein beta 3 subunit gene polymorphisms in Greeks with irritable bowel syndrome. *Dig Dis Sci* 2011; **56**: 3276-3280 [PMID: 21559741 DOI: 10.1007/s10620-011-1726-7]

35 **van der Schaar PJ**, van Hoboken E, Ludidi S, Masclee AA. Effect of cholecystokinin on rectal motor and sensory function in patients with irritable bowel syndrome and healthy controls. *Colorectal Dis* 2013; **15**: e29-e34 [PMID: 23006632 DOI: 10.1111/codi.12034]

36 **El-Salhy M**, Gundersen D, Hatlebakk JG, Gilja OH, Hausken T. Abnormal rectal endocrine cells in patients with irritable bowel syndrome. *Regul Pept* 2014; **188**: 60-65 [PMID: 24316398 DOI: 10.1007/s10620-010-1169-6]

37 **Park SY**, Rew JS, Lee SM, Ki HS, Lee KR, Cheo JH, Kim HI, Noh DY, Joo YE, Kim HS, Choi SK. Association of CCK(1) Receptor Gene Polymorphisms and Irritable Bowel Syndrome in Korean. *J Neurogastroenterol Motil* 2010; **16**: 71-76 [PMID: 20535329 DOI: 10.5056/jnm.2010.16.1.71]

38 **Cheung CK**, Wu JC. Genetic polymorphism in pathogenesis of irritable bowel syndrome. *World J Gastroenterol* 2014; **20**: 17693-17698 [PMID: 25548468 DOI: 10.3748/wjg.v20.i47.17693]

39 **Wouters MM**, Lambrechts D, Knapp M, Cleynen I, Whorwell P, Agréus L, Dlugosz A, Schmidt PT, Halfvarson J, Simrén M, Ohlsson B, Karling P, Van Wanrooy S, Mondelaers S, Vermeire S, Lindberg G, Spiller R, Dukes G, D'Amato M, Boeckxstaens G. Genetic variants in CDC42 and NXPH1 as susceptibility factors for constipation and diarrhoea predominant irritable bowel syndrome. *Gut* 2014; **63**: 1103-1111 [PMID: 24041540 DOI: 10.1136/gutjnl-2013-304570]

40 **Pojoga C**, Stănculete MF. Biopsychosocial Approach of Gastrointestinal Disorders. *Clujul Med* 2014; **87**: 95-97 [PMID: 26528005 DOI: 10.15386/cjmed-289]

41 **Sperber AD**, Dumitrascu D, Fukudo S, Gerson C, Ghoshal UC, Gwee KA, Hungin APS, Kang JY, Minhu C, Schmulson M, Bolotin A, Friger M, Freud T, Whitehead W. The global prevalence of IBS in adults remains elusive due to the heterogeneity of studies: a Rome Foundation working team literature review. *Gut* 2017; **66**: 1075-1082 [PMID: 26818616 DOI: 10.1136/gutjnl-2015-311240]

42 **Gazouli M**, Wouters MM, Kapur-Pojskić L, Bengtson MB, Friedman E, Nikčević G, Demetriou CA, Mulak A, Santos J, Niesler B. Lessons learned--resolving the enigma of genetic factors in IBS. *Nat Rev Gastroenterol Hepatol* 2016; **13**: 77-87 [PMID: 26726033 DOI: 10.1038/nrgastro.2015.206]

43 **Boeckxstaens GE**, Drug V, Dumitrascu D, Farmer AD, Hammer J, Hausken T, Niesler B, Pohl D, Pojskic L, Polster A, Simren M, Goebel-Stengel M, Van Oudenhove L, Vassallo M, Wensaas KA, Aziz Q, Houghton LA; COST Action BM1106 GENIEUR members. Phenotyping of subjects for large scale studies on patients with IBS. *Neurogastroenterol Motil* 2016; **28**: 1134-1147 [PMID: 27319981 DOI: 10.1111/nmo.12886]

44 **Kapeller J**, Houghton LA, Mönnikes H, Walstab J, Möller D, Bönisch H, Burwinkel B, Autschbach F, Funke B, Lasitschka F, Gassler N, Fischer C, Whorwell PJ, Atkinson W, Fell C, Büchner KJ, Schmidtmann M, van der Voort I, Wisser AS, Berg T, Rappold G, Niesler B. First evidence for an association of a functional variant in the microRNA-510 target site of the serotonin receptor-type 3E gene with diarrhea predominant irritable bowel syndrome. *Hum Mol Genet* 2008; **17**: 2967-2977 [PMID: 18614545 DOI: 10.1093/hmg/ddn195]

45 **Kilpatrick LA**, Labus JS, Coveleskie K, Hammer C, Rappold G, Tillisch K, Bueller JA, Suyenobu B, Jarcho JM, McRoberts JA, Niesler B, Mayer EA. The HTR3A polymorphism c. -42C> T is associated with amygdala responsiveness in patients with irritable bowel syndrome. *Gastroenterology* 2011; **140**: 1943-1951 [PMID: 21420406 DOI: 10.1053/j.gastro.2011.03.011]

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SYSTEMATIC REVIEWS

Systematic literature review of the antitumor effect of octreotide in neuroendocrine tumors

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Abstract

AIM

To provide a comprehensive examination of the existing evidence of the antitumor effect of long-acting octreotide in neuroendocrine tumors (NETs).

METHODS

A systematic literature review of clinical trials and observational studies was conducted in PubMed, EMBASE, and Cochrane through January 18, 2017. Conference abstracts for 2015 and 2016 from 5 scientific meetings were also searched.

RESULTS

Of 41 articles/abstracts identified, 13 unique studies compared octreotide with active or no treatment. Two of the 13 studies were clinical trials; the remaining were observational studies. The phase 3 Placebo-Controlled, Double-Blind, Prospective, Randomized Study of the Effect of Octreotide long-acting repeatable (LAR) in the Control of Tumor Growth in Patients with Metastatic

Neuroendocrine Midgut Tumors clinical trial showed that long-acting octreotide significantly prolonged time to tumor progression compared with placebo in patients with functionally active and inactive metastatic midgut NETs; no statistically significant difference in overall survival (OS) was observed, possibly due to the crossover of placebo patients to octreotide. Retrospective observational studies found that long-acting octreotide use was associated with significantly longer OS than no octreotide use for patients with distant metastases although not for those with local/regional disease.

CONCLUSION

The clinical trial and observational studies with informative evidence support long-acting octreotide's antitumor effect on time to tumor progression and OS. This review showed the rarity of existing studies assessing octreotide's antitumor effect and recommends that future research is warranted.

Key words: Neuroendocrine tumors; Antitumor effect; Octreotide; Overall survival; Progression-free survival

Core tip: This review comprehensively summarizes the existing clinical trial and observational studies that have assessed long-acting octreotide's tumor control effect. The comparative studies of relatively large sample size support long-acting octreotide's antitumor effect on time to tumor progression and overall survival. This review shows the rarity of existing studies assessing octreotide's antitumor effect; future research is warranted.

Barrows SM, Cai B, Copley-Merriman C, Wright KR, Castro CV, Soufi-Mahjoubi R. Systematic literature review of the antitumor effect of octreotide in neuroendocrine tumors. *World J Meta-Anal* 2018; 6(2): 9-20 Available from: URL: http://www.wjgnet.com/2308-3840/full/v6/i2/9.htm DOI: http://dx.doi.org/10.13105/wjma.v6.i2.9

INTRODUCTION

Neuroendocrine tumors (NETs) are rare, slow-growing neoplasms[1] that most commonly arise in the gastrointestinal tract, lung, and pancreas[2]. Neuroendocrine tumors account for only 0.5% of all malignancies, with an estimated annual incidence of approximately 2/100000[3]. However, the incidence has been rising, possibly due to increased awareness, improved diagnosis, or evolving definition[3]. Using Surveillance, Epidemiology, and End Results (SEER) data, Dasari *et al*[4] reported an increase in the annual age-adjusted incidence from 1973 (1.09/100000) to 2012 (6.98/100000). Survival for patients with NETs depends on the stage at diagnosis and site of disease. Dasari *et al*[4] reported a median overall survival (OS) for all stages of NETs of 9.3 years. The authors observed that patients with localized NETs had a better median OS (> 30 years) compared with patients with regional NETS (10.2 years) and distant NETs (12 mo). Further, Dasari *et al*[4] observed improvements in OS over time: survival for patients with NETs who were diagnosed in 2009-2012 improved compared with patients with NETs who were diagnosed in 2000-2004 [hazard ratio (HR): 0.79; 95%CI: 0.73-0.85]. Over these same 3 time intervals (2000-2004; 2005-2008; and 2009-2012), improvements in OS were observed for patients with distant-stage gastrointestinal (GI) NETs (HR: 0.71; 95%CI: 0.62-0.81) and in distant-stage pancreatic NETs (HR: 0.56; 95%CI: 0.44-0.70)[4].

Current National Comprehensive Cancer Network (NCCN) guidelines for treatment of NETs recommend the use of somatostatin analogs (SSAs; octreotide and lanreotide) as first-line treatment in patients with advanced NETs[5]. Additional treatment options are based on patient symptoms and the primary tumor location. For patients with unresectable NETs of the pancreas and/or distant metastases who have progressed on treatment with an SSA, octreotide or lanreotide may be continued in combination with everolimus, sunitinib, or chemotherapy[5]. In a review of the available clinical data of octreotide and lanreotide as antitumor agents, the authors concluded that both octreotide and lanreotide have comparable antitumor efficacy and, thus, are interchangeable[6].

Although approved in the United States only for carcinoid symptom (severe diarrhea/flushing episodes) control and not for tumor control, octreotide has been a mainstay of NET therapy for nearly 3 decades[7]. In December 2014, another SSA, lanreotide, was approved for tumor control (*i.e.*, "treatment of patients with unresectable, well- or moderately differentiated, locally advanced or metastatic gastroenteropancreatic NETs to improve progression-free survival")[8].

Sidéris *et al*[2] reviewed literature indexed in MEDLINE (search dates not provided) that identified prospective clinical trials examining the antitumor effects of octreotide and lanreotide in patients with NETs[2]. Six studies published from 1991-1999 showed that 15%[9] to 85.7%[10] of patients with advanced NETs reported stable disease with subcutaneous octreotide[2]. Sidéris *et al*[2] reported that, after the introduction of long-acting octreotide, overall stable disease was observed in 26% to 87.5%[11] of patients with advanced, functioning or nonfunctioning NETs. In those studies that reported partial response, up to 31% of patients receiving subcutaneous octreotide[9] and up to 11% of patients receiving long-acting octreotide experienced a partial response[2].

Broder *et al*[1] conducted a systematic review of literature indexed in PubMed and Cochrane from 1998-2012 to evaluate the efficacy and safety of long-acting octreotide used at higher doses than the United States Food and Drug Administration-approved 30 mg per month. The authors concluded that a summary of

the data suggests a trend supporting the use of high-dose, long-acting octreotide for control of symptoms and limited data supporting the use of high-dose, long-acting octreotide for control of tumor progression in patients with NETs[1]. Several publications provided expert opinion statements that mostly endorsed the use of above-label doses of long-acting octreotide for patients with symptom or tumor progression when lower doses were inadequate to control disease[1]. Most expert opinion publications suggested that higher doses should be used in cases where there is tumor progression or lack of symptom control on lower doses[1]. A recently published review of escalated-dose SSAs in gastroenteropancreatic NETs by Chan *et al*[12] also found evidence of octreotide's antiproliferative effects.

These previous reviews focused on escalated doses of SSAs[1,12] and clinical trials of the antitumor effect of SSAs[2]. At the time of the current review, no systematic reviews summarizing both clinical trial and observational data had been published. Our objective was to provide a systematic and comprehensive review of the existing evidence on the antitumor effect of long-acting octreotide in NETs regardless of dosing and to broaden the search to include real-world evidence and clinical trials.

MATERIALS AND METHODS

We searched PubMed, EMBASE, and the Cochrane Library databases for prospective and retrospective studies evaluating the antitumor effect of octreotide in patients with NETs. Additional studies not published in the peer-reviewed literature were identified by searching online conference abstracts of 5 professional societies: American Society of Clinical Oncology (ASCO), European Society of Medical Oncology, North American Neuroendocrine Tumor Society, European Neuroendocrine Tumor Society, and ASCO-Gastrointestinal Cancers Symposium.

To supplement our search, we also reviewed the bibliographic reference lists of relevant systematic review articles.

The search terms for the medical library databases included Medical Subject Heading, Emtree, and free-text terms of "neuroendocrine tumors," "neuroendocrine neoplasms," "neuroendocrine malignanc*," "neuroendocrine carcinoma," "carcinoid," "octreotide," "Sandostatin," "SMS 201-995," various terms to identify specific antitumor and antiproliferative effect and other outcomes of interest, and terms to identify observational studies, randomized controlled trials, clinical trials, and case series studies. The search was limited to English language studies of humans but had no date limit.

Two independent reviewers screened the titles and abstracts according to predefined inclusion and exclusion criteria (Supplementary Table 1). Full-text articles of selected records were obtained, and the 2 independent reviewers further screened each article according to the same predefined inclusion and exclusion criteria.

RESULTS

The literature database search identified 745 unique records. Six additional articles were identified following a review of the bibliographic reference lists of relevant systematic review articles. One additional abstract was identified from the search of professional societies and associated conferences. A total of 41 publications met inclusion criteria (Figure 1). Of the 41 publications, 20 reported comparative analyses, and 21 reported single-arm studies.

Comparative studies

A total of 20 publications of comparative analyses were identified based on 13 unique studies. Two of the 13 studies were clinical trials, and the remaining were observational studies.

Comparative studies of octreotide vs placebo or no treatment

Four publications reported results of comparisons of long-acting octreotide to placebo or no treatment. This included 2 prospective analyses of the Placebo-Controlled, Double-Blind, Prospective, Randomized Study of the Effect of Octreotide long-acting repeatable (LAR) in the Control of Tumor Growth in Patients with Metastatic Neuroendocrine Midgut Tumors (PROMID) study[13,14] and 2 retrospective analyses of the SEER database[15,16].

Evidence of an antitumor effect of octreotide in patients with midgut NETs was confirmed with the results of the phase 3 PROMID study[13]. Long-acting octreotide significantly lengthened time to tumor progression compared with placebo in patients with functionally active and inactive metastatic midgut NETs. Median time to tumor progression for the long-acting octreotide (n = 42) group was 14.3 mo compared with 6 mo in the placebo (n = 43) group (HR: 0.34; 95%CI: 0.20-0.59; P = 0.000072). After 6 mo of treatment, stable disease was observed in 66.7% of octreotide-treated patients *vs* 37.2% of patients in the placebo group[13]. Rinke *et al*[14] reported final results of median OS for long-acting octreotide and placebo in the PROMID trial as 84.7 and 83.7 mo, respectively (HR: 0.83; 95%CI: 0.47-1.46; P = 0.51). There was a trend toward improved survival in patients with low hepatic tumor load receiving long-acting octreotide *vs* placebo (median not reached *vs* 87.2 mo; HR: 0.59; 95%CI: 0.29-1.2; P = 0.142). Crossover of the majority of placebo patients to long-acting octreotide may have confounded the OS data[14].

Two long-term retrospective analyses were conducted using overlapping periods within the SEER-Medicare database[15,16]. Patients were at least 65 years of age and had functional and nonfunctional NETs originating at

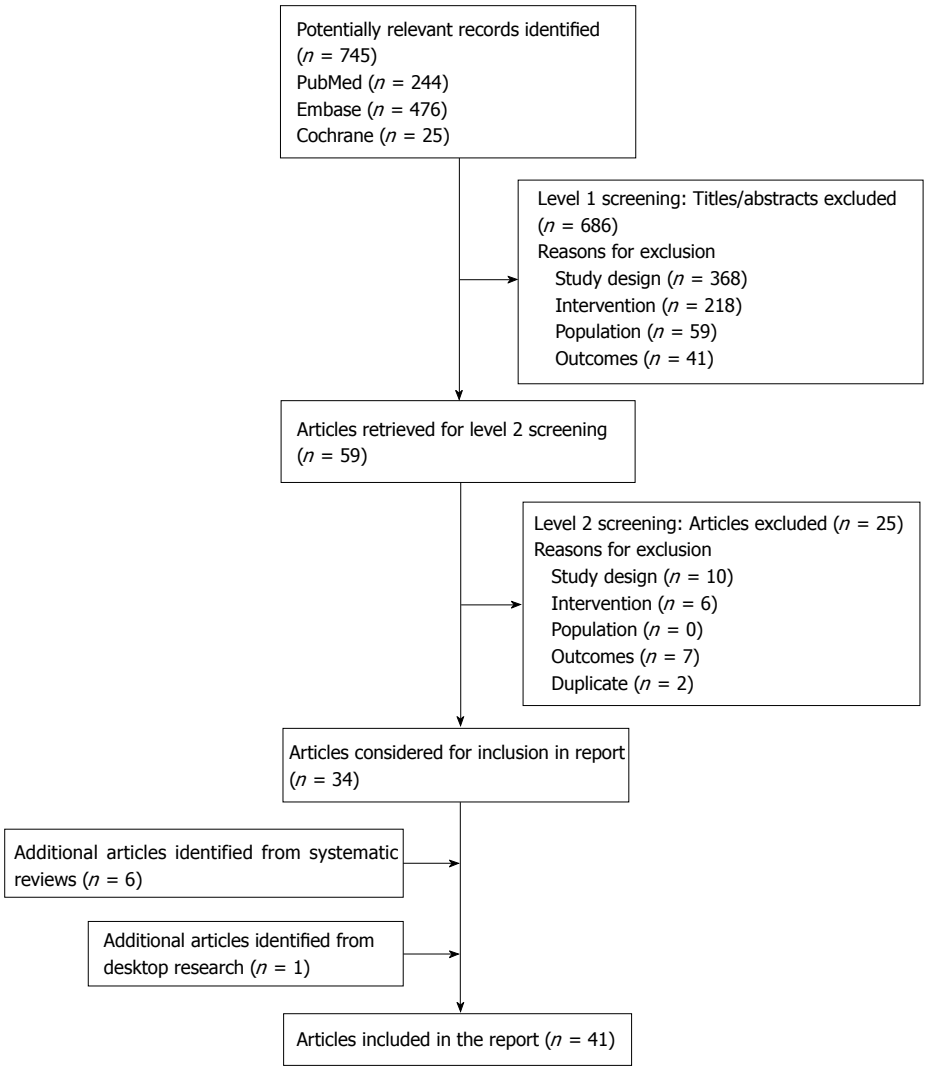

Figure 1 PRISMA diagram. PRISMA diagram describes the search, screening, and selection processes applied in this systematic literature review.

varying sites. In both studies, long-acting octreotide (dose not defined) was compared with no octreotide treatment[15,16]. Shen *et al*[16] (cohort entry July 1999-December 2009 with follow-up through December 2011) reported that in patients with functional or nonfunctional NETs and distant-stage disease, median OS for patients who started long-acting octreotide within 12 mo of diagnosis was significantly longer (35.22 mo; 95%CI: 27.96-47.77) than for those who did not receive octreotide (19.15 mo, 95%CI: 16.36-22.80; HR: 0.68, 95%CI: 0.554-0.840; $P < 0.001$)[16]. In patients with local/regional disease, median OS was 64.85 mo in patients who received long-acting octreotide compared with 104.97 mo in patients who did not receive octreotide (HR: 1.253; 95%CI: 0.928-1.692; $P = 0.1415$)[16]. Shen *et al*[16] further reported a significant survival benefit in the subgroups of patients with distant-stage disease with (HR: 0.65; $P = 0.003$) and without carcinoid syndrome (HR: 0.55; $P = 0.002$). In the analysis reported by Shen *et al*[15] (cohort entry July 1999-December 2007 with follow-up through December 2009), patients with functional NETs and distant-stage disease who received long-acting octreotide within 6 mo of diagnosis had significantly longer median OS (2.11 years; 95%CI: 1.73-2.84 years) than patients who did not receive long-acting octreotide (1.25 years; 95%CI: 0.72-1.71 years; $P = 0.002$). No significant survival benefit was found among the group of patients with NETs of local/regional stage. Further analysis demonstrated that long-acting octreotide was associated with significant improvement in 5-year survival for the subgroup of patients with distant-stage disease (HR: 0.61; 95%CI: 0.47-0.79; $P \leqslant 0.001$). There was no significant benefit observed for patients with local/regional stage disease (HR: 0.88; 95%CI: 0.57-1.36; $P = 0.563$).

Comparative studies comparing different dosing regimens

Five studies involving 28 to 392 patients compared different dose regimens or frequency of dosing for long-acting octreotide; of these, 1 study was prospective and 4 were retrospective. A prospective study examined retrospective data of patients who had been treated

with standard-dose long-acting octreotide 30 mg every 28 d and compared it with the same patients after switching to long-acting octreotide 30 mg every 21 days. The shorter dose interval (*i.e.*, 21 d *vs* 28 d) showed a longer time to tumor progression (30 mo *vs* 9 mo, $P < 0.0001$), and 93% of patients on the 21 d schedule had stable disease[17]. Using the SEER-Medicare database, Shen *et al*[18] estimated the 5-year survival of patients with NETs who received long-acting octreotide within 12 mo of diagnosis. Multivariate analysis showed that, compared with a medium long-acting octreotide dose (21-30 mg), a low dose (≤ 20 mg) was associated with significantly worse survival (HR: 2.000; $P = 0.0011$), whereas a high initial dose (> 30 mg) did not show additional survival benefits over that observed with a medium dose (HR: 1.094; $P = 0.7193$)[18].

Anthony and Vinik[19] (2011) conducted a retrospective medical record review comparing different doses of long-acting octreotide (20, 30, 40, and 60 mg) in which 390 patients were evaluated for tumor response. At the most common dose (long-acting octreotide 30 mg), the rates of complete and partial tumor response were 1% and 8%, respectively. Logistic regression analysis identified no statistically significant correlation between tumor progression and response and the patient's dose, sex, carcinoid syndrome status, and change in dose[19].

In another retrospective medical record review ($n = 54$), Chadha *et al*[20] reported that, in patients with gastroenteropancreatic NETs (GEP-NET), conventional long-acting octreotide (20-30 mg) demonstrated lower estimated 1-year survival and time to any other intervention *vs* high-dose (median, 40 mg) long-acting octreotide, but the results were not statistically significant[20].

In a retrospective medical record review conducted in 43 patients with pancreatic NETs treated with long-acting octreotide, a comparison of low-dose (≤ 20 mg) *vs* medium-dose (30 mg) long-acting octreotide showed longer time to tumor progression for medium dose though, again, the results were not statistically significant[21].

Comparative studies assessing long-acting octreotide monotherapy vs another monotherapy treatment

Three studies (1 prospective, 1 retrospective, and 1 indirect comparison) in 30-110 patients compared octreotide monotherapy to another monotherapy treatment[22-24]. A phase 3 trial comparing long-acting pasireotide 60 mg every 28 d ($n = 53$) and long-acting octreotide 40 mg every 28 d ($n = 57$) showed a higher tumor control rate and median PFS for long-acting pasireotide than long-acting octreotide, but the results were not statistically significant[22]. In a small retrospective medical record review, octreotide 30 mg ($n = 20$) *vs* lanreotide 120 mg ($n = 10$) showed no statistically significant differences in median PFS or 5-year OS[23]. Median PFS was 11.1 mo (95%CI: 7.0-15.2) in the octreotide group *vs* 10.1 mo (95%CI: 4.3-17.0) in the lanreotide group ($P = 0.769$). Five-year OS was 65.6% (95%CI: 29.4-86.6) in the octreotide group and 87.5% (95%CI: 38.7-98.1) in the lanreotide group ($P = 0.864$)[23]. In a study that indirectly compared 182 mo of treatment with recombinant interferon α-2c (2×10^6 IU/m^2 daily; $n = 17$) and octreotide (3×200 μg subcutaneous daily; $n = 16$), stable disease was reported in 85.7% of patients treated with recombinant interferon α-2c and 37.5% of patients treated with subcutaneous octreotide[24].

Table 1 summarizes the 21 publications comparing long-acting octreotide with no treatment or placebo, different octreotide doses, or other monotherapy treatment.

Comparative studies assessing octreotide combination therapy vs octreotide monotherapy

Eight prospective studies compared octreotide combination therapy with octreotide monotherapy[25-32]. Five of the studies were based on the RADIANT-2 study[25-29], 2 studies compared subcutaneous octreotide plus interferon α with subcutaneous octreotide monotherapy[30,31], and 1 study compared long-acting octreotide plus ^{177}Lu-Dotatate with long-acting octreotide monotherapy[32]. The results did not inform the main question of interest for this study (*i.e.*, an antitumor effect of octreotide). Further information pertaining to these studies can be found in the online supplement and Supplementary Table 2.

Single-arm studies

A total of 21 studies were identified as single-arm studies that evaluated the antitumor effect of octreotide. The studies had varying sample sizes ($n = 7$-254), tumor types, and octreotide dosing regimens. The results did not inform the main question of interest for this study (*i.e.*, an antitumor effect of octreotide). Further information pertaining to these studies can be found in the online supplement and Supplementary Table 3[9-11,33-50].

DISCUSSION

This review identified existing clinical trials and observational studies that assessed the antitumor effect of octreotide in patients with NETs. The strongest clinical trial evidence supporting octreotide's antitumor effect was in the phase 3, randomized, placebo-controlled PROMID clinical trial; compared with placebo, long-acting octreotide demonstrated significantly longer time to tumor progression in patients with functionally active or inactive metastatic midgut NETs with or without secretory symptoms[13]. OS did not appear to be significantly different between the two arms, possibly because most patients in the placebo group crossed over to the octreotide arm. There was a trend toward

Table 1 Comparative studies (with no treatment/placebo, different doses, or monotherapy treatment as the comparator)

First author	Study design	No. of patients	Tumor type	Treatment and dose	Treatment duration/ observation period	TTP, mo	SD, %	PR, %	OS, mo	5-yr survival	PFS, mo
Octreotide *vs* placebo or no treatment											
Rinke *et al*[13] (2009) PROMID study	RCT	85	Well-differentiated, advanced NET with midgut or unknown origin. Functional and nonfunctional	LA OCT 30 mg every 28 d (n = 42) *vs* PBO (n = 43)	Patients enrolled between March 2001 and Jan 2008; followed until June 2008	Median OCT: 14.3 *vs* PBO: 6.0 HR: 0.34; 95%CI: 0.20-0.59; P = 0.000072	At 6 mo: OCT: 66.7 *vs* PBO: 37.2 (P = 0.0079)	At 6 mo: 1 in each group	Interim analysis: Median: OCT: Not reached (> 77.4) *vs* PBO: 73.7 HR: 0.81; 95%CI: 0.30-2.18; P = 0.77	-	-
Rinke *et al*[14] (2017) PROMID study	RCT	85	Well-differentiated, advanced NET with midgut or unknown origin. Functional and nonfunctional	LA OCT 30 mg every 28 d (n = 42) *vs* PBO (n = 43)	Patients enrolled between March 2001 and Jan 2008; followed until May 2014	-	-	-	Final analysis Median: OCT: 84.7 *vs* PBO: 83.7 HR: 0.83; 95%CI: 0.47-1.46; P = 0.51	-	-
Shen *et al*[15]1 (2014)	RWE	1291	Distant and local/regional disease; well, moderately, and unknown differentiated tumors with various origin Functional NETs	LA OCT (dose not defined) *vs* no LA OCT	Cohort entry: July 1999-Dec 2007 Follow-up through Dec 2009	-	-	-	Distant stage: OCT: 2.11 y *vs* no OCT: 1.25 y; P = 0.002 Local/regional stage: "no significant survival benefit"	Distant-stage: HR: 0.61; 95%CI: 0.47-0.79; $P \leqslant$ 0.001 Local/regional stage: HR: 0.88; 95%CI: 0.57-1.36; P = 0.563	-
Shen *et al*[16]2 (2015)	RWE	6940	Distant and local/regional disease; well, moderately, and unknown. differentiated tumors with various origin Functional and nonfunctional NETs	LA OCT and no LA OCT distant stage (n = 1176) local/regional stage (n = 5764)	Cohort entry: Jan 1999-Dec 2009 Follow-up through Dec 2011	-	-	-	Distant stage: OCT: 35.22 *vs* no OCT: 19.15 HR: 0.68; 95%CI: 0.554-0.840; P < 0.001 Local/regional stage: OCT: 64.85 *vs* no OCT: 104.97 HR: 1.253; 95%CI: 0.928-1.692; P = 0.1415	-	-

Anthony and Vinik[19] (2011)	RWE	392	Tumor pathology: NR. Localized/ metastatic/ unknown disease with various origin	Without carcinoid syndrome (n = 106) With carcinoid syndrome (n = 260) Carcinoid syndrome after initiation of treatment (n = 24) Overall population initial dose: LA OCT 20 mg: 49% LA OCT 30 mg: 39%	NR	-	57 (any dose)	6 (any dose)	-	-	-
Chadha *et al*[20] (2009)	RWE	54	Tumor pathology: NR Metastatic disease with GEP origin	OCT conventional dose (20 or 30 mg every month; n = 24) OCT high dose (40-90 mg^3; n = 30)	Median follow-up, mo: OCT conventional dose: 35.8 OCT high dose: 44.1	-	-	-	1 yr OS: 0.77 *vs* 0.88; P = 0.4777)	-	-
Ferolla *et al*[17] (2012)	CT	28	Well differentiated functional and nonfunctional NET with various origin	LA OCT 30 mg every 28 d (n = 28) LA OCT 30 mg every 21 d (n = 28)	NR	-	- *vs* 93	- *vs* 7	-	-	-
Jann *et al*[21] (2013)	RWE	43	G1/G2/ Unknown KI-67 index Functional and nonfunctional metastatic tumors with pancreas origin	LA OCT 30 mg (n = 19) LA OCT ⩽ 20 mg (n = 16) OCT (dose unknown(n = 8)	Median follow-up: 58 mo	-	37 (any dose)	5 (any dose)	Median, 98	-	-

Shen *et al*[18][2] (2016)	RWE	222	Well, moderately, or poorly differentiated Functional and nonfunctional distant-stage NETs with various origin	LA OCT every 28 d by dose: ⩽ 20 mg (*n* = 81) 21-30 mg (*n* = 82) > 30 mg (*n* = 59)	Cohort entry: Jan 1999-Dec 2009 Follow-up through Dec 2011	-	-	-	⩽ 20 mg: 20.8 21-30 mg: 32.6 > 30 mg: 36.3 ⩽ 20 mg *vs* 21-30 mg: HR: 2.000; 95%CI: 1.318-3.035; *P* = 0.0011 > 30 mg *vs* 21-30 mg: HR: 1.094; 95%CI: 0.671-1.788; *P* = 0.7193	-	-
Octreotide monotherapy *vs* another monotherapy											
Bongiovanni *et al*[23] (2017)	RWE	30	Well or moderately differentiated locally advanced/ metastatic tumors with lung origin	LA OCT 30 mg every 28 d (*n* = 20) LAN 120 mg every 28 d (*n* = 10)	Median follow-up , 40 mo	-	-	-	-	65.6% *vs* 87.5% (*P* = 0.864)	11.1 *vs* 10.1 (*P* = 0.769)
Creutzfeldt *et al*[24] (1991)	CT	33	Tumor pathology: NR Metastatic gastrointestinal tumors	IFN-α2c (2 × 106 IU/m2 daily; *n* = 17) OCT (200 μg 3 times daily, 500 μg 3 times daily if tumor progressed; *n* = 16)	NR	-	85.7 *vs* 37.5 Comparison NR	-	-	-	-
Wolin *et al*[22] (2015)	RCT	110	Well, moderate, or poorly differentiated Locally advanced/ metastatic tumors with various origin	PAS LAR 60 mg every 28 d (*n* = 53) LA OCT 40 mg every 28 d (*n* = 57)	NR	-	70.6 *vs* 73.1 Comparison NR	2.0 *vs* 1.9 Comparison NR	-	-	-

[1]Sample restricted to patients who either never received treatment with long-acting octreotide or who received it within 6 mo of index date; [2]Sample restricted to patients who either never received treatment with long-acting octreotide or who received it within 12 mo of index date; [3]Median: 40 mg. "-" indicates that data were not reported. CI: Confidence interval; CT: Controlled trial; GEP: Gastroenteropancreatic; HR: Hazard ratio; IFN: Interferon; LAN: Lanreotide; LA: Long-acting; NET: Neuroendocrine tumor; NR: Not reported; OCT: Octreotide; OS: Overall survival; PAS: Pasireotide; PBO: Placebo; PD: Progressive disease; PFS: Progression-free survival; PR: Partial response; PROMID: Placebo-Controlled, Double-Blind, Prospective, Randomized Study of the Effect of Octreotide LAR (long-acting release) in the Control of Tumor Growth in Patients with Metastatic Neuroendocrine Midgut Tumors; RCT: Randomized controlled trial; RWE: Real-world evidence; SD: Stable disease; TTP: Time to tumor progression.

improved OS in patients with a low hepatic tumor load receiving long-acting octreotide compared with placebo[14].

Three retrospective analyses of overlapping periods of SEER-Medicare data provide the strongest retrospective evidence for an antitumor effect of long-acting octreotide, indicating that use of long-acting octreotide was associated with significantly longer OS than no octreotide treatment among patients with distant metastases of various origin, and that standard dosing (21-30 mg) seems to be associated with better OS than low dose (≤ 20 mg)[15,16,18]. These studies provided unique and valuable real-world evidence in the association between long-acting octreotide and OS in tumors of various origin. In the real-world clinical setting, accurate assessment of tumor progression may be challenging due to the rare use of a consistent tumor progression measure (*e.g.*, Response Evaluation Criteria in Solid Tumors, or RECIST); therefore, OS, defined by verified mortality data, is a more consistent study endpoint. SEER-Medicare data allow the long-term follow-up from diagnosis to mortality (longest time period: cohort entry July 1999 to December 2009 with follow-up through December 2011), regardless of changes in providers or health plans. The large sample size and long-term follow-up complements the limitation of clinical studies, which are typically small in sample size and are not powered to assess OS, especially when subject to majority crossover between arms. However, these observational studies assessed only the Medicare population, which is not nationally representative of the NET population, and the crossover between the octreotide and placebo groups may underestimate the OS difference[16].

Current NCCN guidelines for treatment of NETs recommend the use of SSAs (octreotide or lanreotide) as first-line treatment in patients with advanced NETs. Additional subsequent-line therapy options are based on patient symptoms and tumor location (*e.g.*, GI, lung, thymus, pancreas). For patients with unresectable NETs of the pancreas and/or distant metastases who have progressed on treatment with an SSA, octreotide or lanreotide may be continued in combination with everolimus, sunitinib, or chemotherapy[5]. In addition, octreotide and lanreotide have been shown to have comparable antitumor efficacy and thus can be considered interchangeable in regard to antitumor activity[6].

This study adds to previous reviews published in 2012[2], 2015[1], and 2017[12] on this topic by broadening the search in multiple databases and not restricting by dose level, study type (*i.e.*, clinical trial or retrospective study), or date of publication. This review suggests that data from the PROMID trial, combined with real-world effectiveness data[15,16,18], support an antitumor effect of octreotide in NETs, thereby fulfilling an unmet need. The strength of this review lies in its comprehensive search, review, and synthesis of the findings, as well as its rigorous methodology.

Many of the studies included in our review exhibit limitations, including small sample sizes, the absence of a comparative arm, and crossover study designs. Additional studies with large sample sizes and a control arm that does not include octreotide are needed to confirm octreotide's antitumor effect. In addition, future studies should include patients with NETs of various origins.

This study systematically provides the most comprehensive review, to our knowledge, on the clinical trial and retrospective studies that have assessed octreotide's antitumor effect. The clinical trial and observational studies with larger sample sizes support the antitumor effect of long-acting octreotide on time to tumor progression and OS. Most existing studies in this area feature small sample sizes or were not designed to comparatively assess octreotide's antitumor effect. This review identified the rarity of existing studies assessing octreotide's antitumor effect and the need for further research using larger sample sizes and well-controlled study designs.

ARTICLE HIGHLIGHTS

Research background

Neuroendocrine tumors (NETs) are rare, slow-growing neoplasms that most commonly arise in the gastrointestinal tract, lung, and pancreas. Although approved in the United States only for carcinoid symptom (severe diarrhea/flushing episodes) control and not for tumor control, octreotide has been a mainstay of NET therapy for nearly 3 decades.

Research motivation

Previous literature reviews focused on escalated doses of somatostatin analogs (SSAs) and clinical trials of the antitumor effect of SSAs. At the time of the current review, no systematic reviews summarizing both clinical trial and observational data had been published.

Research objective

The objective of this literature review was to provide a systematic and comprehensive examination of the existing evidence of the antitumor effect of long-acting octreotide in NETs regardless of dosing and to broaden the search to include real-world evidence and clinical trials.

Research methods

A systematic literature review of clinical trials and observational studies was conducted in PubMed, EMBASE, and Cochrane through January 18, 2017. Conference abstracts for 2015 and 2016 from 5 scientific meetings were also searched. To supplement the search, the bibliographic reference lists of relevant systematic review articles were also reviewed. Two independent reviewers screened the titles and abstracts according to predefined inclusion and exclusion criteria. Full-text articles of selected records were obtained, and the 2 independent reviewers further screened each article according to the same predefined inclusion and exclusion criteria.

Research results

Of 41 articles/abstracts identified, 13 unique studies compared octreotide with active or no treatment. Two of the 13 studies were clinical trials; the remaining were observational studies. The phase 3 Placebo-Controlled, Double-Blind, Prospective, Randomized Study of the Effect of Octreotide long-acting repeatable (LAR) in the Control of Tumor Growth in Patients with Metastatic Neuroendocrine Midgut Tumors clinical trial showed that long-

acting octreotide significantly prolonged time to tumor progression compared with placebo in patients with functionally active and inactive metastatic midgut NETs; no statistically significant difference in overall survival (OS) was observed, possibly due to the crossover of placebo patients to octreotide. Retrospective observational studies found that long-acting octreotide use was associated with significantly longer OS than no octreotide use for patients with distant metastases although not for those with local/regional disease.

Research conclusion

The clinical trial and observational studies with informative evidence support long-acting octreotide's antitumor effect on time to tumor progression and OS. This review showed the rarity of existing studies assessing octreotide's antitumor effect and recommends that future research is warranted.

REFERENCES

1 **Broder MS**, Beenhouwer D, Strosberg JR, Neary MP, Cherepanov D. Gastrointestinal neuroendocrine tumors treated with high dose octreotide-LAR: a systematic literature review. *World J Gastroenterol* 2015; **21**: 1945-1955 [PMID: 25684964 DOI: 10.3748/wjg.v21.i6.1945]

2 **Sidéris L**, Dubé P, Rinke A. Antitumor effects of somatostatin analogs in neuroendocrine tumors. *Oncologist* 2012; **17**: 747-755 [PMID: 22628056 DOI: 10.1634/theoncologist.2011-0458]

3 **Taal BG**, Visser O. Epidemiology of neuroendocrine tumours. *Neuroendocrinology* 2004; **80** Suppl 1: 3-7 [PMID: 15477707 DOI: 10.1159/000080731]

4 **Dasari A**, Shen C, Halperin D, Zhao B, Zhou S, Xu Y, Shih T, Yao JC. Trends in the Incidence, Prevalence, and Survival Outcomes in Patients With Neuroendocrine Tumors in the United States. *JAMA Oncol* 2017; **3**: 1335-1342 [PMID: 28448665 DOI: 10.1001/jamaoncol.2017.0589]

5 **National Comprehensive Cancer Network**. Clinical practice guidelines: neuroendocrine tumors. Version 3.2017. National Comprehensive Cancer Network, 2017. Available from: URL: https://www.nccn.org/professionals/physician_gls/pdf/neuroendocrine.pdf

6 **Enzler T**, Fojo T. Long-acting somatostatin analogues in the treatment of unresectable/metastatic neuroendocrine tumors. *Semin Oncol* 2017; **44**: 141-156 [PMID: 28923213 DOI: 10.1053/j.seminoncol.2017.07.001]

7 **Sandostatin LAR package insert**. United States: Novartis; July 1, 2016. Available from: URL: https://www.pharma.us.novartis.com/sites/www.pharma.us.novartis.com/files/sandostatin_lar.pdf

8 **Somatuline depot (lanreotide) injection prescribing information**. Ipsen Biopharmaceuticals, Inc.; December 2014. Available from: URL: https://www.accessdata.fda.gov/drugsatfda_docs/label/2014/022074s011lbl.pdf

9 **Anthony L**, Johnson D, Hande K, Shaff M, Winn S, Krozely M, Oates J. Somatostatin analogue phase I trials in neuroendocrine neoplasms. *Acta Oncol* 1993; **32**: 217-223 [PMID: 7686764 DOI: 10.3109/02841869309083915]

10 **Angeletti S**, Corleto VD, Schillaci O, Moretti A, Panzuto F, Annibale B, Delle Fave G. Single dose of octreotide stabilize metastatic gastro-entero-pancreatic endocrine tumours. *Ital J Gastroenterol Hepatol* 1999; **31**: 23-27 [PMID: 10091100]

11 **Tomassetti P**, Migliori M, Corinaldesi R, Gullo L. Treatment of gastroenteropancreatic neuroendocrine tumours with octreotide LAR. *Aliment Pharmacol Ther* 2000; **14**: 557-560 [PMID: 10792118 DOI: 10.1046/j.1365-2036.2000.00738.x]

12 **Chan DL**, Ferone D, Albertelli M, Pavlakis N, Segelov E, Singh S. Escalated-dose somatostatin analogues for antiproliferative effect in GEPNETS: a systematic review. *Endocrine* 2017; **57**: 366-375 [PMID: 28726183 DOI: 10.1007/s12020-017-1360-z]

13 **Rinke A**, Müller HH, Schade-Brittinger C, Klose KJ, Barth P, Wied M, Mayer C, Aminossadati B, Pape UF, Bläker M, Harder J, Arnold C, Gress T, Arnold R; PROMID Study Group. Placebo-controlled, double-blind, prospective, randomized study on the effect of octreotide LAR in the control of tumor growth in patients with metastatic neuroendocrine midgut tumors: a report from the PROMID Study Group. *J Clin Oncol* 2009; **27**: 4656-4663 [PMID: 19704057 DOI: 10.1200/JCO.2009.22.8510]

14 **Rinke A**, Wittenberg M, Schade-Brittinger C, Aminossadati B, Ronicke E, Gress TM, Müller HH, Arnold R; PROMID Study Group. Placebo-Controlled, Double-Blind, Prospective, Randomized Study on the Effect of Octreotide LAR in the Control of Tumor Growth in Patients with Metastatic Neuroendocrine Midgut Tumors (PROMID): Results of Long-Term Survival. *Neuroendocrinology* 2017; **104**: 26-32 [PMID: 26731483 DOI: 10.1159/000443612]

15 **Shen C**, Shih YC, Xu Y, Yao JC. Octreotide long-acting repeatable use among elderly patients with carcinoid syndrome and survival outcomes: a population-based analysis. *Cancer* 2014; **120**: 2039-2049 [PMID: 24676892 DOI: 10.1002/cncr.28653]

16 **Shen C**, Shih YC, Xu Y, Yao JC. Octreotide long-acting repeatable among elderly patients with neuroendocrine tumors: a survival analysis of SEER-Medicare data. *Cancer Epidemiol Biomarkers Prev* 2015; **24**: 1656-1665 [PMID: 26315553 DOI: 10.1158/1055-9965.EPI-15-0336]

17 **Ferolla P**, Faggiano A, Grimaldi F, Ferone D, Scarpelli G, Ramundo V, Severino R, Bellucci MC, Camera LM, Lombardi G, Angeletti G, Colao A. Shortened interval of long-acting octreotide administration is effective in patients with well-differentiated neuroendocrine carcinomas in progression on standard doses. *J Endocrinol Invest* 2012; **35**: 326-331 [PMID: 21757992 DOI: 10.3275/7869]

18 **Shen C**, Xu Y, Dasari A, Shih YC, Yao JC. Octreotide LAR Dosage and Survival Among Elderly Patients With Distant-Stage Neuroendocrine Tumors. *Oncologist* 2016; **21**: 308-313 [PMID: 26911407 DOI: 10.1634/theoncologist.2015-0381]

19 **Anthony L**, Vinik AI. Evaluating the characteristics and the management of patients with neuroendocrine tumors receiving octreotide LAR during a 6-year period. *Pancreas* 2011; **40**: 987-994 [PMID: 21697761 DOI: 10.1097/MPA.0b013e31821f66b4]

20 **Chadha MK**, Lombardo J, Mashtare T, Wilding GE, Litwin A, Raczyk C, Gibbs JF, Kuvshinoff B, Javle MM, Iyer RV. High-dose octreotide acetate for management of gastroenteropancreatic neuroendocrine tumors. *Anticancer Res* 2009; **29**: 4127-4130 [PMID: 19846960]

21 **Jann H**, Denecke T, Koch M, Pape UF, Wiedenmann B, Pavel M. Impact of octreotide long-acting release on tumour growth control as a first-line treatment in neuroendocrine tumours of pancreatic origin. *Neuroendocrinology* 2013; **98**: 137-143 [PMID: 23797176 DOI: 10.1159/000353785]

22 **Wolin EM**, Jarzab B, Eriksson B, Walter T, Toumpanakis C, Morse MA, Tomassetti P, Weber MM, Fogelman DR, Ramage J, Poon D, Gadbaw B, Li J, Pasieka JL, Mahamat A, Swahn F, Newell-Price J, Mansoor W, Öberg K. Phase III study of pasireotide long-acting release in patients with metastatic neuroendocrine tumors and carcinoid symptoms refractory to available somatostatin analogues. *Drug Des Devel Ther* 2015; **9**: 5075-5086 [PMID: 26366058 DOI: 10.2147/DDDT.S84177]

23 **Bongiovanni A**, Recine F, Riva N, Foca F, Liverani C, Mercatali L, Nicolini S, Pieri F, Amadori D, Ibrahim T. Outcome Analysis of First-line Somatostatin Analog Treatment in Metastatic Pulmonary Neuroendocrine Tumors and Prognostic Significance of 18 FDG-PET/CT. *Clin Lung Cancer* 2017; **18**: 415-420 [PMID: 27956089 DOI: 10.1016/j.cllc.2016.11.004]

24 **Creutzfeldt W**, Bartsch HH, Jacubaschke U, Stöckmann F. Treatment of gastrointestinal endocrine tumours with interferon-alpha and octreotide. *Acta Oncol* 1991; **30**: 529-535 [PMID: 1713038 DOI: 10.3109/02841869109092413]

25 **Pavel ME**, Hainsworth JD, Baudin E, Peeters M, Horsch D, Winkler RE, Klimovsky J, Lebwohl D, Jehl V, Wolin EM,

Oberg K, Van Cutsem E, Yao JC, Group R-S. Everolimus plus octreotide long-acting repeatable for the treatment of advanced neuroendocrine tumours associated with carcinoid syndrome (RADIANT-2): a randomised, placebo-controlled, phase 3 study. *Lancet* 2011; **378**: 2005-2012 [DOI: 10.1016/S0140-6736(11)61742-X]

26 **Anthony LB**, Pavel ME, Hainsworth JD, Kvols LK, Segal S, Hörsch D, Van Cutsem E, Öberg K, Yao JC. Impact of Previous Somatostatin Analogue Use on the Activity of Everolimus in Patients with Advanced Neuroendocrine Tumors: Analysis from the Phase III RADIANT-2 Trial. *Neuroendocrinology* 2015; **102**: 18-25 [PMID: 25824001 DOI: 10.1159/000381715]

27 **Castellano D**, Bajetta E, Panneerselvam A, Saletan S, Kocha W, O'Dorisio T, Anthony LB, Hobday T; RADIANT-2 Study Group. Everolimus plus octreotide long-acting repeatable in patients with colorectal neuroendocrine tumors: a subgroup analysis of the phase III RADIANT-2 study. *Oncologist* 2013; **18**: 46-53 [PMID: 23263288 DOI: 10.1634/theoncologist.2012-0263]

28 **Fazio N**, Granberg D, Grossman A, Saletan S, Klimovsky J, Panneerselvam A, Wolin EM. Everolimus plus octreotide long-acting repeatable in patients with advanced lung neuroendocrine tumors: analysis of the phase 3, randomized, placebo-controlled RADIANT-2 study. *Chest* 2013; **143**: 955-962 [PMID: 23187897 DOI: 10.1378/chest.12-1108]

29 **Strosberg JR**, Yao JC, Bajetta E, Aout M, Bakker B, Hainsworth JD, Ruszniewski PB, Van Cutsem E, Öberg K, Pavel ME. Efficacy of octreotide long-acting repeatable in neuroendocrine tumors: RADIANT-2 placebo arm post hoc analysis. *Endocr Relat Cancer* 2015; **22**: 933-940 [PMID: 26373569 DOI: 10.1530/ERC-15-0314]

30 **Arnold R**, Rinke A, Klose KJ, Muller HH, Wied M, Zamzow K, Schmidt C, Schade-Brittinger C, Barth P, Moll R, Koller M, Unterhalt M, Hiddemann W, Schmidt-Lauber M, Pavel M, Arnold CN. Octreotide versus octreotide plus interferon-alpha in endocrine gastroenteropancreatic tumors: a randomized trial. *Clin Gastroenterol Hepatol* 2005; **3**: 761-771 [PMID: 16234004 DOI: 10.1016/S1542-3565(05)00481-7]

31 **Kölby L**, Persson G, Franzén S, Ahrén B. Randomized clinical trial of the effect of interferon alpha on survival in patients with disseminated midgut carcinoid tumours. *Br J Surg* 2003; **90**: 687-693 [PMID: 12808615 DOI: 10.1002/bjs.4149]

32 **Strosberg J**, El-Haddad G, Wolin E, Hendifar A, Yao J, Chasen B, Mittra E, Kunz PL, Kulke MH, Jacene H, Bushnell D, O'Dorisio TM, Baum RP, Kulkarni HR, Caplin M, Lebtahi R, Hobday T, Delpassand E, Van Cutsem E, Benson A, Srirajaskanthan R, Pavel M, Mora J, Berlin J, Grande E, Reed N, Seregni E, Öberg K, Lopera Sierra M, Santoro P, Thevenet T, Erion JL, Ruszniewski P, Kwekkeboom D, Krenning E; NETTER-1 Trial Investigators. Phase 3 Trial of 177 Lu-Dotatate for Midgut Neuroendocrine Tumors. *N Engl J Med* 2017; **376**: 125-135 [PMID: 28076709 DOI: 10.1056/NEJMoa1607427]

33 **Butturini G**, Bettini R, Missiaglia E, Mantovani W, Dalai I, Capelli P, Ferdeghini M, Pederzoli P, Scarpa A, Falconi M. Predictive factors of efficacy of the somatostatin analogue octreotide as first line therapy for advanced pancreatic endocrine carcinoma. *Endocr Relat Cancer* 2006; **13**: 1213-1221 [PMID: 17158766 DOI: 10.1677/erc.1.01200]

34 **Shojamanesh H**, Gibril F, Louie A, Ojeaburu JV, Bashir S, Abou-Saif A, Jensen RT. Prospective study of the antitumor efficacy of long-term octreotide treatment in patients with progressive metastatic gastrinoma. *Cancer* 2002; **94**: 331-343 [PMID: 11900219 DOI: 10.1002/cncr.10195]

35 **Welin SV**, Janson ET, Sundin A, Stridsberg M, Lavenius E, Granberg D, Skogseid B, Oberg KE, Eriksson BK. High-dose treatment with a long-acting somatostatin analogue in patients with advanced midgut carcinoid tumours. *Eur J Endocrinol* 2004; **151**: 107-112 [PMID: 15248829 DOI: 10.1530/eje.0.1510107]

36 **Saglam S**, Hacisahinogullari H, Ozturk N, Kapran Y, Gulluoglu M, Turkmen C, Adalet I, Orhan Bilge A, Cem Balci N. Outcomes of first-line long-acting octreotide treatment in non-functional, advanced gastroenteropancreatic neuroendocrine tumors. *J BUON* 2015; **20**: 1201-1205 [PMID: 26537065]

37 **Arnold R**, Trautmann ME, Creutzfeldt W, Benning R, Benning M, Neuhaus C, Jürgensen R, Stein K, Schäfer H, Bruns C, Dennler HJ. Somatostatin analogue octreotide and inhibition of tumour growth in metastatic endocrine gastroenteropancreatic tumours. *Gut* 1996; **38**: 430-438 [PMID: 8675099 DOI: 10.1136/gut.38.3.430]

38 **Arnold R**, Benning R, Neuhaus C, Rolwage M, Trautmann ME. Gastroenteropancreatic endocrine tumours: effect of Sandostatin on tumour growth. The German Sandostatin Study Group. *Digestion* 1993; **54** Suppl 1: 72-75 [PMID: 8359572 DOI: 10.1159/000201081]

39 **Bajetta E**, Catena L, Procopio G, Bichisao E, Ferrari L, Della Torre S, De Dosso S, Iacobelli S, Buzzoni R, Mariani L, Rosai J. Is the new WHO classification of neuroendocrine tumours useful for selecting an appropriate treatment? *Ann Oncol* 2005; **16**: 1374-1380 [PMID: 15939719 DOI: 10.1093/annonc/mdi258]

40 **Chung MH**, Pisegna J, Spirt M, Giuliano AE, Ye W, Ramming KP, Bilchik AJ. Hepatic cytoreduction followed by a novel long-acting somatostatin analog: a paradigm for intractable neuroendocrine tumors metastatic to the liver. *Surgery* 2001; **130**: 954-962 [PMID: 11742323 DOI: 10.1067/msy.2001.118388]

41 **di Bartolomeo M**, Bajetta E, Buzzoni R, Mariani L, Carnaghi C, Somma L, Zilembo N, di Leo A. Clinical efficacy of octreotide in the treatment of metastatic neuroendocrine tumors. A study by the Italian Trials in Medical Oncology Group. *Cancer* 1996; **77**: 402-408 [DOI: 10.1002/(SICI)1097-0142(19960115)77:2<402::AID-CNCR25>3.0.CO;2-4]

42 **Janson ET**, Oberg K. Long-term management of the carcinoid syndrome. Treatment with octreotide alone and in combination with alpha-interferon. *Acta Oncol* 1993; **32**: 225-229 [PMID: 7686765 DOI: 10.3109/02841869309083916]

43 **Panzuto F**, Di Fonzo M, Iannicelli E, Sciuto R, Maini CL, Capurso G, Milione M, Cattaruzza MS, Falconi M, David V, Ziparo V, Pederzoli P, Bordi C, Delle Fave G. Long-term clinical outcome of somatostatin analogues for treatment of progressive, metastatic, well-differentiated entero-pancreatic endocrine carcinoma. *Ann Oncol* 2006; **17**: 461-466 [PMID: 16364959 DOI: 10.1093/annonc/mdj113]

44 **Ricci S**, Antonuzzo A, Galli L, Ferdeghini M, Bodei L, Orlandini C, Conte PF. Octreotide acetate long-acting release in patients with metastatic neuroendocrine tumors pretreated with lanreotide. *Ann Oncol* 2000; **11**: 1127-1130 [PMID: 11061606 DOI: 10.1023/A:1008383132024]

45 **Saltz L**, Trochanowski B, Buckley M, Heffernan B, Niedzwiecki D, Tao Y, Kelsen D. Octreotide as an antineoplastic agent in the treatment of functional and nonfunctional neuroendocrine tumors. *Cancer* 1993; **72**: 244-248 [DOI: 10.1002/1097-0142(19930701)72:1<244::AID-CNCR2820720143>3.0.CO;2-Q]

46 **Al-Efraij K**, Aljama MA, Kennecke HF. Association of dose escalation of octreotide long-acting release on clinical symptoms and tumor markers and response among patients with neuroendocrine tumors. *Cancer Med* 2015; **4**: 864-870 [PMID: 25727756 DOI: 10.1002/cam4.435]

47 **Laskaratos FM**, Walker M, Naik K, Maragkoudakis E, Oikonomopoulos N, Grant L, Meyer T, Caplin M, Toumpanakis C. Predictive factors of antiproliferative activity of octreotide LAR as first-line therapy for advanced neuroendocrine tumours. *Br J Cancer* 2016; **115**: 1321-1327 [PMID: 27811856 DOI: 10.1038/bjc.2016.349]

48 **Öberg K**, Norheim I, Theodorsson E. Treatment of malignant mid-gut carcinoid tumours with a long-acting somatostatin analogue octreotide. *Acta Oncol* 1991; **30**: 503-507 [PMID: 27811856 DOI: 10.1038/bjc.2016.349]

49 **Ramundo V**, Del Prete M, Marotta V, Marciello F, Camera L, Napolitano V, De Luca L, Circelli L, Colantuoni V, Di Sarno A, Carratu AC, de Luca di Roseto C, Colao A, Faggiano A. Impact of long-acting octreotide in patients with early-stage MEN1-related duodeno-pancreatic neuroendocrine tumours. *Clin Endocrinol* (Oxf)

2014; **80**: 850-855 [PMID: 24443791 DOI: 10.1111/cen.12411]
50 **Wang Y**, Wang W, Jin K, Fang C, Lin Y, Xue L, Feng S, Zhou Z, Shao C, Chen M, Yu X, Chen J. Somatostatin receptor expression indicates improved prognosis in gastroenteropancreatic neuroendocrine neoplasm, and octreotide long-acting release is effective and safe in Chinese patients with advanced gastroenteropancreatic neuroendocrine tumors. *Oncol Lett* 2017; **13**: 1165-1174 [PMID: 28454229 DOI: 10.3892/ol.2017.5591]

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EDITORIAL

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EDITORIAL

Improving the conduct of meta-analyses of observational studies

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Abstract

The author, who has published numerous meta-analyses of epidemiological studies, particularly on tobacco, comments on various aspects of their content. While such meta-analyses, even when well conducted, are more difficult to draw inferences from than are meta-analyses of clinical trials, they allow greater insight into an association than do simple qualitative reviews. This editorial starts with a discussion of some problems relating to hypothesis definition. These include the definition of the outcome, the exposure and the population to be considered, as well as the study inclusion and exclusion criteria. Under literature searching, the author argues against restriction to studies published in peer-reviewed journals, emphasising the fact that relevant data may be available from other sources. Problems of identifying studies and double counting are discussed, as are various issues in regard to data entry. The need to check published effect estimates is emphasised, and techniques to calculate estimates from material provided in the source publication are described. Once the data have been collected and an overall effect estimate obtained, tests for heterogeneity should be conducted in relation to different study characteristics. Though some meta-analysts recommend classifying studies by an overall index of study quality, the author prefers to separately investigate heterogeneity by those factors which contribute to the assessment of quality. Reasons why an association may not actually reflect a true causal relationship are also discussed, with the editorial describing techniques for investigating the relevance of confounding, and referring to problems resulting from misclassification of key variables. Misclassification of disease, exposure and confounding variables can all produce a spurious association, as can misclassification of the variable used to determine whether an individual can enter the study, and the author points to techniques to adjust for this. Issues relating to publication bias and the interpretation of "statistically significant" results are also discussed. The editorial should give the reader insight into the difficulties of producing a good meta-analysis.

Key words: Hypothesis definition; Literature searching; Heterogeneity; Publication bias; Misclassification; Confounding; Meta-analysis

Core tip: The author has published many meta-analyses of epidemiological studies, particularly on smoking, and the editorial comments on various aspects of their conduct. Areas covered include the definition of the hypothesis to be tested, literature searching and data entry, as well as methods to test for heterogeneity and investigate such issues as confounding, misclassification and publication bias. The need for well conducted meta-analyses and the difficulty in determining whether a "statistically significant" association is actually indicative of a causal relationship are discussed. The editorial should be helpful to readers inexperienced with the conduct of meta-analyses.

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INTRODUCTION

Meta-analyses were originally designed to combine data from randomized controlled trials, with the Quality of Reporting of Meta-analyses statement[1] describing how the quality of such meta-analyses could be improved. Provided the trials which were being combined were of sufficiently similar design, and involved the same exposures and outcomes there was little difficulty in interpreting the overall effect estimate. Such meta-analyses clearly had greater power to detect relationships than had the individual studies being combined.

For many years attempts to summarize evidence on an association from multiple observational epidemiological studies were based on qualitative reviews. These reviews typically summarized the results of each study in a paragraph or two, and then attempted to draw an overall conclusion. International Agency for Research on Cancer monographs was often qualitative and it is sometimes difficult to see the process by which the overall conclusion had been reached.

Bringing meta-analysis techniques to the field of observational studies seemed attractive in that it provided some sort of quantitative overall assessment, but there was initially considerable concern about the validity of combining results from studies using different designs and methods, and conducted in different countries and time periods where the nature of the exposure may have varied. While there is clearly some element of truth in the criticism that one should not combine "apples and oranges", it became clear over the years that well-conducted meta-analyses can be extremely useful in assisting the judgement as to whether a relationship is a causal one. Particularly where the association is strong is consistently seen in multiple well conducted studies, and there is no source of confounding or bias that materially affects the estimates, one seems to be on safe grounds to conclude that a causal relationship exists.

Over the years, I and my colleagues at P.N. Lee Statistics and Computing Ltd. have conducted a large number of meta-analyses relating to the health effects of tobacco. These consider effects of smoking generally[2-5], different types of cigarette[6-8], quitting[9-12], smokeless tobacco[13-15], Swedish "snus"[16-18] and nicotine replacement therapy[19], as well as effects of parental smoking[20-22] and of environmental tobacco smoke exposure[23-28]. Mainly these meta-analyses relate to outcomes which are 1/0 variables (typically presence or absence of a disease), though some concern continuous outcomes such as forced expiratory volume[29,30] or cholesterol level[31]. While I do not have experience of conducting meta-analyses in other areas, I have also served as a reviewer for numerous meta-analyses submitted to journals and I hope that some of the knowledge I have accumulated will be of interest to others.

This editorial is not intended to describe how meta-analyses should be structured or presented. This is adequately described in the meta-analysis of observational studies in epidemiology proposals[32] and the preferred reporting items for systematic reviews and meta-analyses (PRISMA) statements[33], while the reporting of meta-analysis protocols is well covered by PRISMA-P[34]. Nor is it intended to cover all aspects of conducting a meta-analysis, what follows really being a collection of personal comments of mine on various aspects of meta-analyses of observational studies.

DEFINING THE HYPOTHESIS TO BE TESTED

While some meta-analyses can be quite broad-ranging, relating a number of aspects of exposure of an agent to a number of different outcomes, others may be much more specific. It is important at the outset to clearly define the objectives of the work, and the hypotheses to be tested.

In a simple case, there may be one specific outcome of interest, and the study protocol should make clear what definitions of that outcome are allowed. For some diseases this may cause few problems, but for others this requires thought. In other cases, there may be several related outcomes, or specific subsets of the outcome, which are of interest. For example, in our review of the evidence relating smoking to chronic obstructive pulmonary disease (COPD), chronic bronchitis and emphysema[2], we had to be careful to define what could be regarded as satisfactorily equivalent diseases, since COPD is a relatively recently used term, and we did not wish to exclude relevant older studies. We were also careful to record the basis of definition used in each study (*e.g.*, symptoms reported on a questionnaire, mortality records), so that we could compare effect estimates according to this definition.

Similar considerations apply to the definition of exposure. First, we have to define what the exposure is - for smoking, for example, are we limiting attention to cigarettes, or do we include cigars and pipes? Are we

considering only exposure above a certain minimum level or any exposure? Are we considering ever exposure or current exposure? If we are considering current exposure are we comparing this with non-current exposure or with never exposure? Should we accept those who have ceased exposure very recently as part of the currently exposed group? Should we accept those with only a minimum lifetime exposure among the never exposed group? Often it may be useful to meta-analyse effect estimates for various exposure definitions. However, it is, in principle, a good idea to define in advance the main exposure of interest, to avoid being accused of trying various alternative definitions and then only reporting or emphasising the one that best shows the association of interest.

For both outcome and exposure, a balance has to be struck between using narrow definitions which may seriously limit the number of eligible studies, or allowing broader definitions which will increase the number of studies (and thus the workload and costs) and may hamper interpretation of the results.

In some situations, the hypothesis of interest is to be tested among a subset of the population. For example, when studying the relationship of environmental tobacco smoke exposure to a disease, it is usual to restrict attention to those who have never smoked (as exposure to tobacco smoke constituents from smoking is typically two orders of magnitude higher than from environmental tobacco smoke exposure). Here, one needs to define whether it is acceptable to include results from studies which include those with minimum lifetime cigarette consumption among the definition of never smoking.

One also has to define study inclusion and exclusion criteria. Are we restricting attention to certain study designs, perhaps only considering cohort studies, or certain sub-populations, such as employed persons? Are we excluding studies in children, or in adults who have relevant co-existing diseases or conditions, or who work in high-risk occupations? Are we only interested in studies which provide dose-response results? There are many possibilities depending on the detail of the study protocol. It may be useful to keep a list of those studies where the decision to reject was a marginal one, partly so that this list can be presented, together with the reason for rejection, in a supplementary file to the paper reporting the results of the meta-analysis, and partly so that results from such rejected papers may be included in sensitivity analyses.

LITERATURE SEARCHING

As discussed elsewhere[33] it is necessary to make it absolutely clear exactly what the search criteria used are, so that others can repeat the searches, perhaps at a later date. Whether one limits attention to Medline searches, on the basis that they are quite comprehensive and free, or to studies published in English, to avoid the costs of translation, is up to the researcher. Especially where such restricted searches provide substantial numbers of relevant studies, extending to other literature databases or studies in other languages may add little useful.

It is sometimes suggested that attention should be restricted to studies published in peer-reviewed journals. I disagree with this view for two reasons. Firstly, my personal experience suggests that peer-review is not necessarily a guarantee of quality. Second, it is the quality of the study that matters, so why should one necessarily reject results from a good study published in a journal which is not peer-reviewed?

Similar considerations apply to unpublished data. In my 50 yr as a practising epidemiologist/medical statistician I have accumulated and filed a number of unpublished reports. If they contain relevant data, why should I not use them? On some occasions, the reviewer may be able to add useful material to his review by conducting analyses on public databases. While the methods used will need to be clearly described, perhaps in a supplementary file to the publication presenting the results of the meta-analyses, there seems in principle to be no good reason to exclude such evidence.

IDENTIFICATION OF STUDIES AND DOUBLE COUNTING

Once a set of suitable papers has been identified from the literature search it will be necessary to draw up a list of studies. Some papers will present results from multiple studies, which it is advisable to keep separate in data entry for proper assessment of between-study heterogeneity. More commonly results from some studies will be presented in multiple publications. If one publication clearly supersedes another (*e.g.*, reporting results from 20 rather than 10 year follow-up from a cohort study), the superseded publication can be omitted from the meta-analysis to avoid double-counting. However, if two publications present independent results (*e.g.*, for different sexes or age groups) then they should both be considered in the meta-analyses.

Complete avoidance of overlap may not be the most desirable solution. For example, a national study based on outcomes occurring in, say, 1990 may include some individuals also considered in a study in a smaller region based on outcomes in 1985 to 1995. Similarly one paper may publish results from a study involving cases in 2000 to 2005 while another may publish results from the same study involving cases in 2004 to 2008. In both examples, complete avoidance would require exclusion of one of the studies, whereas, given the minor overlap, it would seem acceptable to include both sets of results.

ENTERING DATA

For complex meta-analysis projects, we have found it useful to have two linked databases, one containing the characteristics of each study and the other the detailed results, typically containing multiple records for each study.

The study database would include a single record per study and contain such information as the relevant publication(s), the sexes considered, the age range of the population, the location of the study and its timing and length of follow-up, the nature of the population studied, any study weaknesses, the definition of the outcome, the numbers of cases and of subjects, the types of controls and matching factors used in case-control studies, the confounding variables studied, and the availability of results for each index of exposure and outcome studied.

Each record on the other database would be linked to the relevant study and refer to a specific effect estimate, recording the comparison made and the results. This record would include such details as the outcome, the sex, details of the exposure considered (including the level of exposure for dose-related indices), the source of the effect estimate (*e.g.*, source publication, with page or table number), the type of effect estimate (*e.g.*, relative risk, hazard ratio or odds ratio for 1/0 outcomes, or means or medians for continuous outcomes), the method of derivation (see below) and the adjustment variables taken into account. It would also include the effect estimate itself and its 95%CI or standard deviation, and the numbers of exposed and unexposed cases and controls (or at risk). It is also advisable to look routinely for errors in reported results. Some years ago I described[35] some simple methods to do this for odds ratios, relative risks and CI, and used these methods to give some examples of seriously erroneous published data, which unless corrected could seriously distort the results of the meta-analyses.

It is also necessary to have a clear set of rules for identifying which effect estimates are to be entered from each study. Is it planned to enter estimates by sex, age or other stratifying variables, or only overall estimates? Are there types of estimate that should not be entered, such as those which are adjusted for symptoms of the disease of interest?

Consideration should also be given to how to handle incompletely reported results. Where studies simply report results as "non-significant", without providing an effect estimate, one at least should mention this in a paper reporting on a meta-analysis. Ideally, an attempt to obtain quantitative estimates from the author should be made.

In many cases the effect estimates can be taken directly from the source publication, but in other cases it will be necessary to calculate them from the material provided (or, if practicable, from raw data supplied by the author of the publication). Often the effect estimates can be calculated using standard methods[36], but there is a situation I commonly come across, where more sophisticated techniques are required. This is where a study presents effect estimates and 95%CI for a range of different exposures (*e.g.*, dose levels) relative to a specific exposure (*e.g.*, unexposed), and one wishes to derive effect estimates and 95%CI for a different comparison (*e.g.*, all exposed *vs* unexposed). Here the important thing to note is that the effect estimates and 95%CI are not independent, as they have a common base, so that the combined estimate cannot be derived by simple meta-analysis of the individual estimates (as would be the situation given simple stratified data, *e.g.*, by age). Fortunately a method to derive an appropriate combined estimate is available[37] and should be used. A method is also available[38] to derive estimates of the increase in effect per unit dose from such a table. Note that when deriving such estimates one will need a method to estimate the mean level of exposure from ranges, including open-ended intervals.

Most of the meta-analyses my colleagues and I have carried out over the years have been based on software we have written ourselves. Simple fixed-effect and random-effects meta-analysis can be programmed quite rapidly in Excel, the relevant methodology being succinctly described in the Appendix to a paper by Fleiss and Gross[39]. More commonly we use software incorporated into the ROELEE system developed by my colleague John Fry. While programming one's own software gives better insight into the methodology, John Fry advises me that 'meta for', the meta-analysis package for R, is a convenient one to use for those who do not wish to get so involved.

STUDY QUALITY

While there are published methods for assessing study quality, such as the Cochran Collaboration Risk of Bias Tool and Effective Public Health Practice Project Quality Assessment Tool[40], or the Newcastle-Ottawa Scale[41] which I have on occasion used, I have always been somewhat sceptical of them, because they seem to be trying to quantify what is essentially multi-dimensional into a single dimension. Even where study quality assessments are made, it is usually advisable to also carry out heterogeneity tests to see how effect estimates vary by those specific study characteristics which contribute to the assessment of quality.

HETEROGENEITY TESTS

Where there are a reasonable number of independent effect estimates to be combined, analyses of heterogeneity should be conducted. If Q is Cochran's heterogeneity statistic, and df is the number of degrees of freedom (one less than the number of estimates combined), then heterogeneity is often expressed by the I^2 statistic which is equal to 100% × (Q − df) / Q. Negative values of I^2 are set equal to zero, so that I^2 lies in the range 0 to 100%, with values of 0% indicating no obvious heterogeneity, larger values indicating increased heterogeneity.

Apart from conducting standard fixed- effect and random-effects meta-analyses (see[39]), a systematic review should also include more detailed tests of heterogeneity, where Q is shown to be statistically significant (at $P < 0.05$) and the number of estimates is sufficiently large (usually at least 10). These more detailed tests would involve separate fixed-effect meta-analyses for different levels of relevant study characteristic - such as sex, location, study type, definition of outcome, definition of exposure, number of confounding variables adjusted for, study size and

Table 1 In a longitudinal study (often referred to as a prospective or cohort study) the data may be expressed

		Predictor variable		
		Exposed	Unexposed	Total
Outcome	Yes	A	B	A + B
	No	C	D	C + D
	Total	A + C	B + D	N

presence of a study weakness. These analyses serve two main purposes - first, to see whether an association seen in the overall meta-analysis is consistently seen in study subsets, and to see whether any factors are the cause of any heterogeneity seen. If a study characteristic has m levels (i = 1, ..., m) and if Q_i is the Cochran heterogeneity statistic for level i, then the statistic $Q^*=Q-\sum_{i=1}^{m}Q_i$ is a test of heterogeneity between levels of the characteristic on m − 1 degrees of freedom. If Q* is close to its degrees of freedom, it implies that the study characteristic explains little or none of the heterogeneity. If, on the other hand, it is close to Q, it suggests that the characteristic is a major determinant of the heterogeneity. Where data permit it is useful to carry out meta-regression analyses in which a model is fitted simultaneously relating the effect estimate to a set of study characteristics. Because of correlation between characteristics, this should give greater insight into which are the important sources of heterogeneity and which are not. Variation in the effect estimate by levels of a study characteristic may arise for different reasons. For example, higher effect estimates in one location may be because of greater exposure to (or differing metabolism of) the exposure of interest by the population there. Or it may be due to differing biases in different situations. For example, higher effect estimates in case-control studies than in cohort studies may suggest that recall bias in case-control studies may be relevant, or for other reasons as described in the next section.

Combining relative risks and odds ratios

Suppose we are studying the relationship or a predictor variable to an outcome, each with two levels. In a longitudinal study (often referred to as a prospective or cohort study) the data may be expressed as in Table 1.

The relationship of outcome to exposure is typically expressed by the relative risk (RR), the ratio of the probability of the outcome given exposure, A / (A + C), to that given no exposure, B / (B + D), or RR = A (B + D)/B (A + C), the variance of its logarithm being given by 1 / A + 1 / B − 1 / (A + C) −1 / (B + D).

In a cross-sectional or case-control study, the data may be similarly expressed, but here the relationship is typically expressed by the odds ratio (OR), the ratio of the odds of the outcome given exposure, A/C, to that given no exposure, B/D or OR = AD/BC, the variance of its logarithm being given by 1 / A + 1 / B + 1 / C + 1 / D.

Where the outcome is relatively rare, it can be shown that RR and OR are very similar. Thus, for example, with A = 10 and B = 20, and a true RR of 2, the OR will be 2.04 when comparing probabilities of 2% and 4%, and even closer to 2 for smaller probabilities. Even comparing 10% and 20% the OR of 2.25 is not that far from 2.

This suggests that when conducting meta-analysis of a reasonably rare outcome, one can combine RRs and ORs without worrying. Where this is not the case, e.g. when comparing 20% and 40% (where the OR is 2.67), this is less valid and it is preferable either to report separate combined results for ORs and RRs, or to try to convert one into the other. This is simple when the data are in the form of a 2 × 2 table, but not possible for adjusted estimates without access to the raw data.

I note that in longitudinal studies, where RRs are in principle more appropriate, ORs are often presented in publications. This is related to the simplicity of adjusting for multiple variables simultaneously using logistic regression analysis.

ADJUSTMENT FOR CONFOUNDING VARIABLES

Especially where the association between the exposure and disease of interest is quite modest, one needs to bear in mind that the association may not be a causal one, and may be due to confounding by one or more variables which are correlated both with the exposure and the disease. Individual study authors are usually well aware of the problem and often present effect estimates adjusted for one or more sets of potential confounders. There are various approaches to investigate confounding in meta-analyses.

One possibility is to extract most-adjusted and least-adjusted effect estimates from each study. Most-adjusted estimates are those estimates reported in the source publication which have been adjusted for the most potential confounding variables, while least-adjusted estimates may include estimates that are totally unadjusted or adjusted only for age. Given these estimates, one can either compare results of meta-analyses based on the alternative estimates, or meta-analyse the ratio of estimates (perhaps using a weight based on the confidence limits of the most-adjusted estimates). Some studies may of course only provide one estimate, and can be excluded from such meta-analyses.

An additional method which may provide insight is to look for heterogeneity of the effect estimate according to the grouped number of confounders adjusted for, or to compare estimates adjusted or unadjusted for specific potential confounding variables.

Where an association substantially reduces following adjustment for confounding, but remains statistically significant, the possibility of bias arises.

Though beyond the scope of most meta-analyses, it is on some occasions worth formally investigating the extent to which effect estimates from meta-analyses may be biased by such uncontrolled confounding. The interested reader may wish to study the techniques used in our systematic review of the relation between environmental

tobacco smoke exposure and lung cancer[23] which concluded that bias due to uncontrolled confounding by four factors (fruit, vegetable and dietary fat consumption, and education) explains a substantial part of the observed association.

Another possibility to be borne in mind is "residual confounding", arising because relevant confounders have not been adjusted for. It is well documented that "misclassification of a confounder" leads to "partial loss of ability to control confounding"[42] while "even misclassification rates as low as 10% can prevent adequate control of confounding"[43]. It has even been noted that if X is an inaccurately measured true cause of disease, and if Y, which is precisely measured but not a cause, is correlated with X, one may incorrectly conclude that Y, not X, is the cause (*e.g.*[44-46]).

MISCLASSIFICATION

Apart from bias arising due to misclassification of confounding variables, bias may also arise because of other forms of misclassification. Random misclassification of the exposure or outcome variable will tend to dilute any relationship, but misclassification may not be random, and can lead to underestimation of the relationship. For example, when studying a relatively weak association of smoking to cancer at one site, the inclusion of some individuals who actually have cancer of a site known to be strongly related to smoking (such as lung cancer) will bias upward the association being studied. Misdiagnosis of lung cancer certainly exists[47-49]. Similarly, upward bias will arise if some of those classified as having the exposure of interest actually have an exposure which is more strongly related to the disease.

While random misclassification of exposure or outcome should not produce an association when no true causal relationship exists, this is certainly not so for random misclassification of the variable used to determine whether an individual should be included in the study. This applies, for example, to the study of the relationship of spousal smoking to lung cancer in never smokers. As I have demonstrated[50,51], the inclusion of some true ever smokers among the reported never smokers, can cause bias. This bias arises because spouses tend to have smoking habits in common, so that the exposed group (with spouses who smoke) are likely to include more misclassified smokers than will the comparison group (with spouses who do not smoke). Because of the very high risk of lung cancer this bias can be substantial, and the interested reader may wish to study the techniques which my colleagues and I used to adjust for misclassification bias[23].

PUBLICATION BIAS

Publication bias occurs if the published data are not representative of all the data that exist on a topic. It is well documented (*e.g.*[52,53]) that positive findings are published more often than negative findings, so meta-analyses of data drawn from the literature tend to overestimate true relationships. Inasmuch as large studies are more likely than small studies to publish their findings regardless of the result, one can compare effect estimates from larger and smaller studies as some sort of test of publication bias. More formal tests are available, but tend to involve assumptions that are difficult to justify. Furthermore, they are based on the published results, and ignore what may be known about unpublished results. What should one conclude if a very large cohort study has published evidence demonstrating a statistically significant relationship between an exposure and various common diseases, but has not reported results relating that exposure to other common diseases? It seems to me quite likely that the authors would have looked at these other diseases, found no significant association, and decided not to publish their findings. The existence of such studies should at least be pointed out in the discussion section of a paper describing a meta-analysis of the exposure to one of these other diseases.

Publication bias can also arise in the meta-analysis of dose-response relationships. It is certainly plausible that authors will be more likely to report dose-response results where there is a strong association in the first place. This can be tested by comparing effect estimates for overall exposure in studies reporting and not reporting dose-response results.

STATISTICAL SIGNIFICANCE

An effect estimate derived from a meta-analysis that is not statistically significant ($P > 0.1$) clearly cannot be interpreted as supporting a true causal relationship. Nor can it rule it out, as one cannot prove a negative, but it can suggest an upper limit to any true effect. Additional studies may clarify the situation, especially where the original meta-analysis had little power, being based on relatively few studies.

On the other side of the coin, a significant association alone does not demonstrate that a true causal effect exists. *P*-values less than 0.05 but greater than 0.1 may be due to chance, and even where the probability is very low, so that chance can be excluded for practical purposes, confounding or bias may be relevant. Before concluding that a causal effect is likely, it is up to the meta-analyst to demonstrate that confounding or bias cannot explain the relationship, which may be difficult, especially where the relationship is weak.

CONCLUSION

Meta-analysis is an interesting subject and quite difficult to do well. If it is done well it can act as an extremely useful tool to aid the epidemiologist in reaching a conclusion. However, it is very important for the meta-analyst to be aware of the limitations of meta-analysis, and of the epidemiological studies on which it is based.

REFERENCES

1 **Moher D**, Cook DJ, Eastwood S, Olkin I, Rennie D, Stroup DF.

Improving the quality of reports of meta-analyses of randomised controlled trials: the QUOROM statement. Quality of Reporting of Meta-analyses. *Lancet* 1999; **354**: 1896-1900 [PMID: 10584742 DOI: 10.1016/S0140-6736(99)04149-5]

2 **Forey BA**, Thornton AJ, Lee PN. Systematic review with meta-analysis of the epidemiological evidence relating smoking to COPD, chronic bronchitis and emphysema. *BMC Pulm Med* 2011; **11**: 36 [PMID: 21672193 DOI: 10.1186/1471-2466-11-36]

3 **Lee PN**, Forey BA, Coombs KJ. Systematic review with meta-analysis of the epidemiological evidence in the 1900s relating smoking to lung cancer. *BMC Cancer* 2012; **12**: 385 [PMID: 22943444 DOI: 10.1186/1471-2407-12-385]

4 **Fry JS,** Lee PN, Forey BA and Coombs KJ. Dose-response relationship of lung cancer to amount smoked, duration and age starting. *World J Meta-Anal* 2013; **1**: 57-77 [DOI: 10.13105/wjma.v1.i2.57]

5 **Lee PN,** Forey BA, Thornton AJ and Coombs KJ. The relationship of cigarette smoking in Japan to lung cancer, COPD, ischemic heart disease and stroke: a systematic review [version 1; referees: awaiting peer review]. *F1000Research* 2018; **7**: 204 [DOI: 10.12688/f1000research.14002.1]

6 **Lee PN**. Lung cancer and type of cigarette smoked. *Inhal Toxicol* 2001; **13**: 951-976 [PMID: 11696868 DOI: 10.1080/08958370175321 0353]

7 **Lee PN**. Systematic review of the epidemiological evidence comparing lung cancer risk in smokers of mentholated and unmentholated cigarettes. *BMC Pulm Med* 2011; **11**: 18 [PMID: 21501470 DOI: 10.1186/1471-2466-11-18]

8 **Lee PN**. Tar level of cigarettes smoked and risk of smoking-related diseases. *Inhal Toxicol* 2018; **30**: 5-18 [PMID: 29488428 DOI: 10.108 0/08958378.2018.1443174]

9 **Lee PN**, Fry JS, Hamling JS. Using the negative exponential distribution to quantitatively review the evidence on how rapidly the excess risk of ischaemic heart disease declines following quitting smoking. *Regul Toxicol Pharmacol* 2012; **64**: 51-67 [PMID: 22728684 DOI: 10.1016/j.yrtph.2012.06.009]

10 **Fry JS**, Lee PN, Forey BA, Coombs KJ. How rapidly does the excess risk of lung cancer decline following quitting smoking? A quantitative review using the negative exponential model. *Regul Toxicol Pharmacol* 2013; **67**: 13-26 [PMID: 23764305 DOI: 10.1016/j.yrtph.2013.06.001]

11 **Lee PN**, Fry JS, Thornton AJ. Estimating the decline in excess risk of cerebrovascular disease following quitting smoking--a systematic review based on the negative exponential model. *Regul Toxicol Pharmacol* 2014; **68**: 85-95 [PMID: 24291341 DOI: 10.1016/j.yrtph.2013.11.013]

12 **Lee PN**, Fry JS, Forey BA. Estimating the decline in excess risk of chronic obstructive pulmonary disease following quitting smoking - a systematic review based on the negative exponential model. *Regul Toxicol Pharmacol* 2014; **68**: 231-239 [PMID: 24361344 DOI: 10.1016/j.yrtph.2013.12.006]

13 **Lee PN**. Circulatory disease and smokeless tobacco in Western populations: a review of the evidence. *Int J Epidemiol* 2007; **36**: 789-804 [PMID: 17591642 DOI: 10.1093/ije/dym039]

14 **Weitkunat R**, Sanders E, Lee PN. Meta-analysis of the relation between European and American smokeless tobacco and oral cancer. *BMC Public Health* 2007; **7**: 334 [PMID: 18005437 DOI: 10.1186/1471-2458-7-334]

15 **Lee PN**, Hamling J. Systematic review of the relation between smokeless tobacco and cancer in Europe and North America. *BMC Med* 2009; **7**: 36 [PMID: 19638245 DOI: 10.1186/1741-7015-7-36]

16 **Lee PN**. Summary of the epidemiological evidence relating snus to health. *Regul Toxicol Pharmacol* 2011; **59**: 197-214 [PMID: 21163315 DOI: 10.1016/j.yrtph.2010.12.002]

17 **Lee PN**. Epidemiological evidence relating snus to health--an updated review based on recent publications. *Harm Reduct J* 2013; **10**: 36 [PMID: 24314326 DOI: 10.1186/1477-7517-10-36]

18 **Lee PN**, Thornton AJ. The relationship of snus use to diabetes and allied conditions. *Regul Toxicol Pharmacol* 2017; **91**: 86-92 [PMID: 29061372 DOI: 10.1016/j.yrtph.2017.10.017]

19 **Lee PN**, Fariss MW. A systematic review of possible serious adverse health effects of nicotine replacement therapy. *Arch Toxicol* 2017; **91**: 1565-1594 [PMID: 27699443 DOI: 10.1007/s00204-016-1856-y]

20 **Thornton AJ**, Lee PN. Parental smoking and risk of childhood cancer: a review of the evidence. *Indoor Built Environ* 1998; **7**: 65-86 [DOI: 10.1159/000024566]

21 **Thornton AJ**, Lee PN. Parental smoking and sudden infant death syndrome: a review of the evidence. *Indoor Built Environ* 1998; **7**: 87-97 [DOI: 10.1159/000024567]

22 **Thornton AJ**, Lee PN. Parental smoking and middle ear disease in children: a review of the evidence. *Indoor Built Environ* 1999; **8**: 21-39 [DOI: 10.1159/000024607]

23 **Lee PN,** Fry JS, Forey B, Hamling JS and Thornton AJ. Environmental tobacco smoke exposure and lung cancer: a systematic review. *World J Meta-Anal* 2016; **4**: 10-43 [DOI: 10.13105/wjma.v4.i2.10]

24 **Lee PN**, Thornton AJ, Hamling JS. Epidemiological evidence on environmental tobacco smoke and cancers other than lung or breast. *Regul Toxicol Pharmacol* 2016; **80**: 134-163 [PMID: 27321059 DOI: 10.1016/j.yrtph.2016.06.012]

25 **Lee PN**, Hamling JS. Environmental tobacco smoke exposure and risk of breast cancer in nonsmoking women. An updated review and meta-analysis. *Inhal Toxicol* 2016; **28**: 431-454 [PMID: 27541291 DOI: 10.1080/08958378.2016.1210701]

26 **Lee PN**, Thornton AJ, Forey BA, Hamling JS. Environmental Tobacco Smoke Exposure and Risk of Stroke in Never Smokers: An Updated Review with Meta-Analysis. *J Stroke Cerebrovasc Dis* 2017; **26**: 204-216 [PMID: 27765554 DOI: 10.1016/j.jstrokecerebrovasdis.2016.09.011]

27 **Lee PN,** Forey BA, Hamling JS and Thornton AJ. Environmental tobacco smoke exposure and heart disease: A systematic review. *World J Meta-Anal* 2017; **5**: 14-40 [DOI: 10.13105/wjma.v5.i2.14]

28 **Lee PN,** Forey BA, Coombs KJ, Hamling JS and Thornton AJ. Epidemiological evidence relating environmental smoke to COPD in lifelong non-smokers: a systematic review [version 1; referees: awaiting peer review]. *F1000Research* 2018; **7**: [DOI: 10.12688/f1000research.13887.1]

29 **Lee PN**, Fry JS. Systematic review of the evidence relating FEV1 decline to giving up smoking. *BMC Med* 2010; **8**: 84 [PMID: 21156048 DOI: 10.1186/1741-7015-8-84]

30 **Fry JS**, Hamling JS, Lee PN. Systematic review with meta-analysis of the epidemiological evidence relating FEV1 decline to lung cancer risk. *BMC Cancer* 2012; **12**: 498 [PMID: 23101666 DOI: 10.1186/1471-2407-12-498]

31 **Forey BA**, Fry JS, Lee PN, Thornton AJ, Coombs KJ. The effect of quitting smoking on HDL-cholesterol - a review based on within-subject changes. *Biomark Res* 2013; **1**: 26 [PMID: 24252691 DOI: 10.1186/2050-7771-1-26]

32 **Stroup DF**, Berlin JA, Morton SC, Olkin I, Williamson GD, Rennie D, Moher D, Becker BJ, Sipe TA, Thacker SB. Meta-analysis of observational studies in epidemiology: a proposal for reporting. Meta-analysis Of Observational Studies in Epidemiology (MOOSE) group. *JAMA* 2000; **283**: 2008-2012 [PMID: 10789670 DOI: 10.1001/jama.283.15.2008]

33 **Liberati A**, Altman DG, Tetzlaff J, Mulrow C, Gøtzsche PC, Ioannidis JP, Clarke M, Devereaux PJ, Kleijnen J, Moher D. The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate healthcare interventions: explanation and elaboration. *BMJ* 2009; **339**: b2700 [PMID: 19622552 DOI: 10.1136/bmj.b2700]

34 **Shamseer L**, Moher D, Clarke M, Ghersi D, Liberati A, Petticrew M, Shekelle P, Stewart LA; PRISMA-P Group. Preferred reporting items for systematic review and meta-analysis protocols (PRISMA-P) 2015: elaboration and explanation. *BMJ* 2015; **350**: g7647 [PMID: 25555855 DOI: 10.1136/bmj.g7647]

35 **Lee PN**. Simple methods for checking for possible errors in reported odds ratios, relative risks and confidence intervals. *Stat Med* 1999; **18**: 1973-1981 [PMID: 10440880 DOI: 10.1002/(SICI)1097-0258(19990815)18:15<1973::AID-SIM166>3.0.CO;2-G]

36 **Gardner MJ**, Altman DG, editors. Statistics with confidence: Confidence intervals and statistical guidelines. London: British Medical Journal 1989; 140

37 **Hamling J**, Lee P, Weitkunat R, Ambühl M. Facilitating meta-analyses by deriving relative effect and precision estimates for alternative comparisons from a set of estimates presented by exposure level or disease category. *Stat Med* 2008; **27**: 954-970 [PMID: 17676579 DOI: 10.1002/sim.3013]

38 **Berlin JA**, Longnecker MP, Greenland S. Meta-analysis of epidemiologic dose-response data. *Epidemiology* 1993; **4**: 218-228 [PMID: 8512986 DOI: 10.1097/00001648-199305000-00005]

39 **Fleiss JL**, Gross AJ. Meta-analysis in epidemiology, with special reference to studies of the association between exposure to environmental tobacco smoke and lung cancer: a critique. *J Clin Epidemiol* 1991; **44**: 127-139 [PMID: 1995774 DOI: 10.1016/0895-4356(91)90261-7]

40 **Armijo-Olivo S**, Stiles CR, Hagen NA, Biondo PD, Cummings GG. Assessment of study quality for systematic reviews: a comparison of the Cochrane Collaboration Risk of Bias Tool and the Effective Public Health Practice Project Quality Assessment Tool: methodological research. *J Eval Clin Pract* 2012; **18**: 12-18 [PMID: 20698919 DOI: 10.1111/j.1365-2753.2010.01516.x]

41 **Wells GA**, Shea B, O'Connell D, Peterson J, Welch V, Losos M and Tugwell P. The Newcastle-Ottawa Scale (NOS) for assessing the quality of nonrandomised studies in meta-analyses. Available from: URL: http://www.ohri.ca/programs/clinical_epidemiology/oxford.asp?status=print

42 **Greenland S**. The effect of misclassification in the presence of covariates. *Am J Epidemiol* 1980; **112**: 564-569 [PMID: 7424903 DOI: 10.1093/oxfordjournals.aje.a113025]

43 **Tzonou A**, Kaldor J, Smith PG, Day NE, Trichopoulos D. Misclassification in case-control studies with two dichotomous risk factors. *Rev Epidemiol Sante Publique* 1986; **34**: 10-17 [PMID: 3726220]

44 **Greenland S**, Robins JM. Confounding and misclassification. *Am J Epidemiol* 1985; **122**: 495-506 [PMID: 4025298 DOI: 10.1093/oxfordjournals.aje.a114131]

45 **Savitz DA**, Barón AE. Estimating and correcting for confounder misclassification. *Am J Epidemiol* 1989; **129**: 1062-1071 [PMID: 2705426 DOI: 10.1093/oxfordjournals.aje.a115210]

46 **Fewell Z**, Davey Smith G, Sterne JA. The impact of residual and unmeasured confounding in epidemiologic studies: a simulation study. *Am J Epidemiol* 2007; **166**: 646-655 [PMID: 17615092 DOI: 10.1093/aje/kwm165]

47 **Lee PN**. Comparison of autopsy, clinical and death certificate diagnosis with particular reference to lung cancer. A review of the published data. *APMIS Suppl* 1994; **45**: 1-42 [PMID: 7811528]

48 **Faccini JM**. The role of histopathology in the evaluation of risk of lung cancer from environmental tobacco smoke. *Exp Pathol* 1989; **37**: 177-180 [PMID: 2637150 DOI: 10.1016/S0232-1513(89)80042-8]

49 **Sterling TD**, Rosenbaum WL, Weinkam JJ. Bias in the attribution of lung cancer as cause of death and its possible consequences for calculating smoking-related risks. *Epidemiology* 1992; **3**: 11-16 [PMID: 1554804 DOI: 10.1097/00001648-199201000-00004]

50 **Lee PN**, Forey BA. Misclassification of smoking habits as a source of bias in the study of environmental tobacco smoke and lung cancer. *Stat Med* 1996; **15**: 581-605 [PMID: 8731002 DOI: 10.1002/(SICI)1097-0258(19960330)15:6<581::AID-SIM182>3.0.CO;2-B]

51 **Lee PN,** Forey BA and Fry JS. Revisiting the association between environmental tobacco smoke exposure and lung cancer risk. III. Adjustment for the biasing effect of misclassification of smoking habits. *Indoor Built Environ* 2001; **10**: 384-398 [DOI: 10.1177/1420326X0101000605]

52 **Sterling TD**. Publication decisions and their possible effects on inferences drawn from tests of significance - or vice versa. *J Am Stat Assoc* 1959; **54**: 30-34 [DOI: 10.2307/2282137]

53 **Easterbrook PJ**, Berlin JA, Gopalan R, Matthews DR. Publication bias in clinical research. *Lancet* 1991; **337**: 867-872 [PMID: 1672966 DOI: 10.1016/0140-6736(91)90201-Y]

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