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Spotlight on Anti-CGRP Monoclonal Antibodies in Migraine: The Clinical Evidence to Date

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Abstract

Migraine, a common neurovascular brain disorder, represents a severe and widespread health problem; along with medication-induced (medication-overuse) headache, it is the third-leading cause of disability worldwide. Currently, its therapeutic management remains unsatisfactory for several reasons; up to 40% of migraineurs are eligible for prophylactic treatment, but there are issues of efficacy, safety, and adherence. In recent years the evidence on the role of calcitonin gene-related peptide (CGRP) in migraine pathophysiology has been consolidated, so new and promising treatments for migraine pain and its possible prevention have been developed. The following review reports the results of the clinical trials conducted so far with each of the new monoclonal antibodies targeting CGRP or its receptor, with particular reference to safety, tolerance, and efficacy in migraine prevention. Moreover, the pharmacological characterization and further developments of each monoclonal antibody are reported, based on current knowledge.

Keywords

monoclonal antibodies, immunotherapy, CGRP, migraine, prophylaxis

A headache is one of the reasons that most often leads a patient to consult a doctor in the clinical neurology setting. Among primary headache disorders, migraine is a common neurovascular brain dysfunction, defined as a recurrent unilateral headache disorder lasting 4–72 hours, characterized by pulsating pain of moderate or severe intensity, associated with nausea and/or photophobia and phonophobia. Approximately, 50% of European adults have an active headache disorder, and about 15% seem to suffer from migraine,[1] which has a higher prevalence in women (16.6%) than in men (7.5%).[2] Currently, migraine is considered a severe and widespread health problem. It is the sixth-leading cause of disability worldwide and the third-leading cause of disability in those younger than 50 years old.[3] In more than 7% of migraineurs, the pain increases in frequency over time, leading to a high-frequency episodic migraine or, even worse, to a chronic disorder, when it occurs during at least 15 days per month for at least 3 months, with approximately 8 episodes per month.[4] The recurrent painful symptoms and the headache-related disability associated with recurrent migraine are two of the best reasons to start prophylactic therapy. Up to 40% of migraineurs are eligible for this treatment, but current therapeutic management is difficult and unsatisfactory because of frequent adverse reactions and poor patient compliance.[5] Various types of prophylactic medications are widely used for high-frequency episodic or chronic migraine, such as anticonvulsants, tricyclic antidepressants, beta-blockers, and calcium channel blockers.[6] However, in a substantial proportion of patients, there are issues of efficacy, safety, adherence, and drug–drug interactions, especially in the case of comorbidities such as cardiovascular and psychiatric diseases.[7] For these reasons, about 1 of 5 migraineurs is forced to suspend pharmacological prophylactic treatment because of adverse events and tolerance issues[8]; meanwhile, 1 of 5 patients is compliant with the prophylactic treatment when it lasts up to a year.[9] It has been estimated that more than 140 million people in the world have chronic migraine,[10] approximately the population of Russia. Most of them are not taking a prophylactic therapy.

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Table 1. Monoclonal Antibodies Targeting the CGRP Pathway in Phase 3 Clinical Studies

Primary Sponsoring Company	INN or Code Name	Molecular Format	Target	Most Advanced Phase	Indications
Alder Biopharmaceuticals	ALD403/ eptinezumab	Humanized IgG_1	CGRP	Phase 3	Migraine prevention
Eli Lilly and Company	LY2951742/ galcanezumab	Humanized IgG_4	CGRP	Phase 3	Migraine and cluster headache prevention
Teva Pharmaceuticals	TEV-48125/ frestanezumab	Humanized IgG_2	CGRP	Phase 3	Migraine prevention
Amgen/Novartis	AMG 334/erenumab	Human IgG_2	CGRP receptor	Phase 3	Migraine prevention

INN, international nonproprietary name; CGRP, calcitonin gene–related peptide.

In the United States, 14 million migraineurs would benefit from preventive therapy; however, it has never been proposed to them.[11] OnabotulinumtoxinA is the only approved treatment by the Food and Drug Administration for chronic migraine.[12,13] Novel and mechanism-based therapies are therefore necessary and should be a focus of continued research to address this tremendous burden.[14,15] From the early hypotheses formulated in 1985,[16] pieces of evidence have reinforced the idea that calcitonin gene-related peptide (CGRP) is a key neuropeptide in migraine pathophysiology, up to the recent evidence of antimigraine effect shown by CGRP receptor blockade.[17] Following the first effective CGRP-receptor antagonists, which are not yet usable for safety reasons, recent attention has been focused on 4 monoclonal antibodies (mAbs) targeting the CGRP pathway, all of which are currently in phase 3 clinical development (Table 1).[18] In this article, we review the current knowledge and state of progress in this area.

CGRP in Migraine Pathophysiology

CGRP is a 37–amino acid neuropeptide, a member of a family of peptides that includes amylin, adrenomedullin, and calcitonin, which actively operate in different parts of the organism and are primarily localized in C unmyelinated sensory fibers.[19] There are 2 isoforms of CGRP, α-CGRP and β-CGRP, which are encoded by alternate splicing of the calcitonin coding gene. These isoforms differ by 3 amino acids and coexist in most neurons. In particular, α-CGRP is mainly expressed in the central nervous system, whereas β-CGRP is predominant in presynaptic terminals of enteric sensory neuronal cells.[20] The 2 isoforms are both complete agonists of the CGRP receptor, which is a membrane heterodimer composed of a 461–amino acid protein with 7 transmembrane domains (calcitonin receptor-like receptor) and a single transmembrane peptide (receptor activity–modifying protein 1).[21] An additional third protein is required to form an optimally functional CGRP receptor, a hydrophilic membrane-associated protein (receptor component protein) able to primarily activate adenylate cyclase and protein kinase A, resulting in the phosphorylation of multiple downstream targets.[21] CGRP is the most potent vasodilator currently known.[22] It mediates a final common pathway in smooth muscle cells, achieving a decrease in intracellular concentration of free calcium ion concentration[23] and subsequent cell relaxation. Moreover, studies have shown that it has a facilitatory role in nociceptive transmission together with other neuromediators, such as substance P and bradykinin.[24,25] This role is especially apparent when it is released from the trigeminal ganglia neurons that innervate the cranial vessels, one of the major sites of neuropeptide synthesis.[26] In recent decades, knowledge of the role of CGRP and its receptor in pain transmission has expanded considerably, mainly in the migraine pathophysiological model. It is now assumed that neuronal dysfunction of the central sensory system[27,28] generates migraine attacks in genetically susceptible individuals. The trigeminal system is clearly involved,[29] and a large quantity of neuropeptides, such as CGRP, are released from terminal nerve endings in the meninges and face. Consequently, the peripheral activities of CGRP and other peptides released, such as substance P (often colocalized in Aδ and C sensory fibers), play a major role in the expression and maintenance of head pain and possibly in other migraine symptoms.[30] The most obvious evidence supporting the role of CGRP in migraine pain showed that increased plasma levels of CGRP were associated with painful syndromes such as migraine and cluster headache.[31] Once the concentration was normalized after the attack ended, CGRP infusion could induce migraine attacks,[32] and triptans were able to normalize the rise in plasma levels during a migraine attack.[33] It was also suggested that increased CGRP plasma levels outside migraine attacks and in the absence of symptomatic treatment could be helpful biomarkers in the diagnosis of chronic migraine.[34] Increasing interest in this neuropeptide has resulted in the development of new and promising treatments for migraine pain

and, possibly, prevention. The first were the effective CGRP-receptor antagonists, for which development was slowed for safety reasons. Thereafter, 4 monoclonal antibodies targeting CGRP or its receptor (anti-CGRP mAbs) were created for migraine prevention with the aim of overcoming the safety issues that affected the above-mentioned CGRP-receptor antagonists.

Pharmacologic Properties

CGRP-receptor antagonists (so-called gepants) were the first drugs developed and tested against the CGRP signaling pathway in humans, where they proved to have adequate efficacy both in acute migraine and in migraine prophylaxis. These agents included BI 44370 TA, which exhibited dose-dependent efficacy in the treatment of acute migraine attacks,[35] and rimegepant, which was superior to placebo at several different doses in the acute treatment of migraine (75, 150, and 300 mg),[36] as well as olcegepant[37] and telcagepant.[38,39] These data provided the first evidence that acting on the CGRP signaling pathway was a new and effective antimigraine approach. However, despite the enormous efforts made, none of these gepants was approved for clinical use because of the risk of liver toxicity after chronic exposure.[40] In the last few years, a new class of biologic drugs able to block the CGRP pathway was developed and subsequently tested in several phase 1 and phase 2 clinical trials, including 3 humanized monoclonal antibodies (mAbs) directed against CGRP (ALD403/eptinezumab, TEV-48125/frestanezumab, and LY2951742/galcanezumab)[41–43] and 1 human mAb targeting the CGRP receptor (AMG 334/erenumab).[44] These macromolecules specifically bound their target (CGRP or CGRP receptor, as shown in Figure 1), with the aim of preventing repeated CGRP-induced trigeminal nociceptive transmission, therefore decreasing headache frequency over time and improving migraine symptoms.[45] The site and mechanism of action of these biological agents are still not completely understood. The mAbs do not cross the blood–brain barrier under physiological conditions[46]; therefore, the first results from the phase 1 and phase 2 clinical trials, in which several patients experienced a complete remission period of several months, seem to suggest a peripheral target of action. The verified poor blood–brain barrier penetration of the effective antimigraine triptans, gepants, and mAb[40] and the evidence that the trigeminal ganglion, like the dura mater, lacks a blood–brain barrier and is freely accessible to circulating compounds[47,48] advanced the hypothesis of a peripheral target in which the mAb can act continuously, as recently confirmed by Schankin et al.[49] The novel radioligand ^{11}C-dihydroergotamine, chemically identical to active dihydroergotamine, was unable to cross the blood–brain barrier in 6 control subjects and 6 migraineurs, ictally or interictally, demonstrating that the blood–brain barrier remains tight during acute glyceryl trinitrate-induced migraine attacks. The pharmacokinetic and pharmacodynamic profiles of mAbs are very different than those of the smaller molecules and/or oral medications used for migraine prophylactic therapy. First, they are administered parenterally because of their large dimensions (approximately 150 000 Da, compared with <1000 Da for gepants), as well as their relatively low permeability through cell membranes and their instability in the gastrointestinal tract. Compared with the small-molecule CGRP-receptor antagonists, which have a half-life ranging in hours, these mAbs have a longer duration of action, with a plasma half-life lasting days or weeks. The extended plasma half-life of monoclonal antibodies allows longer dosing intervals, with a subcutaneous or intravenous administration that is preferably carried out every month or quarterly. This is a suitable characteristic for prophylactic treatments because there is no need to take the drug on a daily basis, thus improving patient compliance.[50] Moreover, the biologic medications are not metabolized by the liver; elimination is primarily via catabolism in smaller peptides and individual amino acids.[51] Therefore, there is a low risk of drug–drug interactions, without the danger of raising creatinine or the hepatic enzymes.[38,52] Information about any relevant pharmacodynamic differences between each of the 4 mAbs is very limited: the first divergences in vitro regarding the CGRP intrinsic binding features of ALD403, TEV-48125, and LY2951742a/LY2951742b were presented at the Annual Scientific Meeting of the American Headache Society in June 2016.[53] There it was hypothesized that these antibodies were not identical, with small differences in targeting the same ligand in association/dissociation rates, which may impact the therapeutic activity of the monoclonal antibody. More in detail, a well-established technology (surface plasmon resonance) was used to characterize the binding of these antibodies to CGRP, and it was noted that LY2951742a/LY2951742b acts as an incomplete antagonist, with rapid target engagement and dissociation. This characteristic can result in significant levels of free CGRP available to engage its receptor and stimulate signaling to a measurable extent ($K_d = 1.4 \times 10^{-5}$ and $K_d = 1.0 \times 10^{-5}$, respectively). In contrast, ALD403 ($K_d = 1.0 \times 10^{-6}$) and TEV-48125 ($K_d = 1.0 \times 10^{-6}$) engaged CGRP differentially, but with undetectable dissociation. This peculiarity may determine some differences in the pharmacological activity of these mAbs, such as in migraine duration, intensity, and analgesic drug efficacy. It was also reported that ALD403 at equivalent circulating concentrations engages and stops CGRP activities twice as rapidly

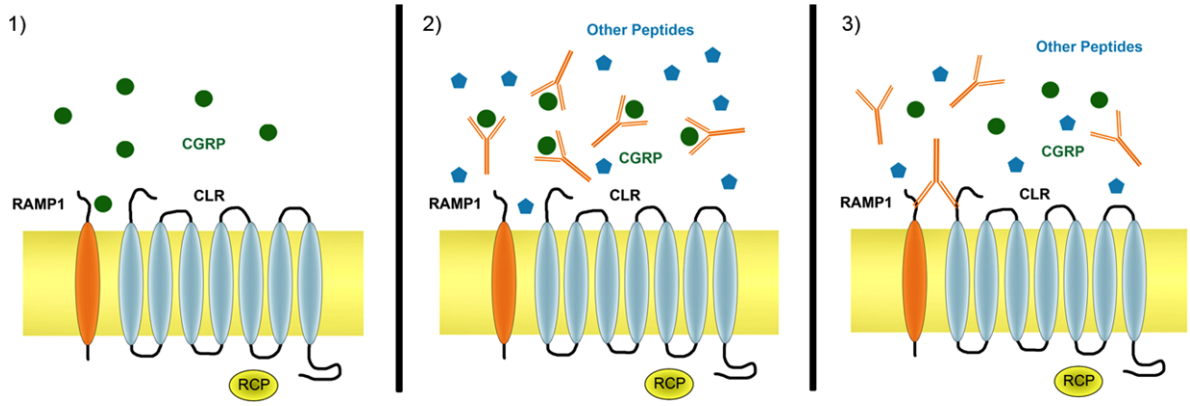

Figure 1. Illustrative representation of calcitonin gene–related peptide (CGRP) activity in the absence and/or in the presence of anti-CGRP mAbs. (1) In normal conditions, CGRP (little green balls) binds usually its receptor. (2) When the CGRP is neutralized by a monoclonal antibody, other peptides (blue pentagons) may potentially interact with the CGRP main receptor. (3) When the CGRP main receptor is blocked by a monoclonal antibody, it is impeded by any receptor interaction. CGRP and other peptides can alternatively still bind other receptors, for which they have affinity.

as TEV-48125. Further evaluation, however, will be required to see if these fine changes have the potential to impact in vivo the effectiveness of an antibody against migraine symptoms.

MAbs in Migraine Prophylactic Therapy: The Evidence to Date

The following paragraphs report the results of the phase 1 and phase 2 clinical trials conducted thus far with each of the 3 anti-CGRP mAbs (ALD403/eptinezumab, LY2951742/galcanezumab, and TEV-48125/frestanezumab) that potently and selectively neutralize both α-CGRP and β-CGRP and the mAb anti-CGRP receptor (AMG 334/erenumab). These data highlight the safety, tolerance, and efficacy of these medications in the prevention of recurrent migraine attacks (the results of phase 2 clinical trials are summarized in Table 2 and Table 3). Moreover, the future developments of each antibody are reported, based on current knowledge.

ALD403/eptinezumab

ALD403/eptinezumab is a fully humanized IgG_1 antibody manufactured using yeast (*Pichia pastoris*) and developed by Alder Biopharmaceuticals, with a half-life of approximately 30 days. The mAb was first implemented in a single-dose, placebo-controlled study (NCT01579383) to determine the safety, tolerability, and pharmacokinetics of the compound administered by intravenous infusion and/or subcutaneous injection. It was completed in April 2013, with 104 healthy men and women between the ages of 18 and 65. The intravenous administration of eptinezumab was associated with a long plasma half-life (~26 days) and linear pharmacokinetics. No sex differences were observed, and there were no pharmacokinetic interactions with subcutaneous sumatriptan.[54] An additional phase 1 study, conducted in Melbourne (Australia), evaluated the pharmacokinetic and pharmacodynamics of eptinezumab administered quarterly via intravenous, subcutaneou,s or intramuscular routes to 60 healthy women and men aged between 18 and 65 years,[55] with good safety and tolerability findings. The next proof of concept was obtained in an exploratory phase 2 trial completed in January 2016 to evaluate the effects of a single dose of eptinezumab administered intravenously to patients with frequent migraine episodes.[41] The study was conducted with 163 men and women between 18 and 55 years old with 5 to 14 migraine days, randomly assigned (1:1) to receive either an intravenous dose of eptinezumab 1000 mg (n = 81) or placebo (n = 82) per 28-day period for up to 24 weeks. Most adverse events were mild to moderate in severity and occurred in 57% of patients in the eptinezumab group and 52% in the placebo group. The most common events were upper respiratory tract infections (6 patients in the placebo group vs 7 patients in the eptinezumab group), urinary tract infections (4 patients vs 1 patient), fatigue (3 vs 3), back pain (4 vs 3), arthralgia (4 vs 1), and nausea (2 vs 3). No infusion reactions were reported during the study. Serious adverse events occurred in 2 patients in the eptinezumab group and 1 patient in the placebo group; these were judged to be unrelated to the treatment. The treatment group proved to have higher response rates for all times and responder rates, with the eptinezumab rate being approximately 20% higher. Moreover, 16% of patients treated with the mAb experienced a 100% reduction in migraine days for the entire study period. At the moment, another phase 2 clinical trial (NCT02275117) is ongoing in the United States, Australia, Georgia, and New Zealand to assess the efficacy, safety, and pharmacokinetics of eptinezumab

Table 2. ALD403 and LY2951742 for Migraine Prevention: Efficacy and Safety Results of Phase 2 Clinical Trials

Antibody	Study Population (Active vs Placebo)	Inclusion Criteria	Treatment Dose and Frequency	Primary Efficacy Outcome (From Baseline)	Change of Primary Efficacy Outcome (Active vs Placebo)	AEs (Number of Patients, Active vs Placebo)	AEs	Patients Positive to Antidrug Antibodies
ALD403/ eptinezumab[41]	163 (81 vs 82)	Episodic migraine	1 g iv, once	MHD at 5–8 weeks	−5.6 vs −4.6 ($P = .0306$)	46 vs 43 (57% vs 52%)	Respiratory and urinary infections, fatigue, and back pain	11 (14%)
ALD403/ eptinezumab — preliminary results	588 (472 vs 116)	Chronic migraine	300 mg iv, once (114 patients)	75% responder rate at 1–12 weeks	38 vs 24 (33% vs 21%) ($P < .05$)	NA	Respiratory infections, nasopharyngitis, and nausea	NA
			100 mg iv, once (118 patients)	75% responder rate at 1–12 weeks	37 vs 24 (31% vs 21%) ($P < .05$)	NA	Dizziness, nausea, and nasopharyngitis	
			30 mg iv, once (117 patients)	75% responder rate at 1–12 weeks	33 vs 24 (28% vs 21%)	NA	Respiratory infections and sinusitis	
			10 mg iv, once (123 patients)	75% responder rate at 1–12 weeks	33 vs 24 (27% vs 21%)	NA	Dizziness, sinusitis, and nausea	
LY2951742/ galcanezumab[43]	218 (107 vs 110)	Episodic migraine	150 mg sc, every 2 weeks	MHD at 9–12 weeks	−4.2 vs −3.0 ($P = .003$)	77 vs 74 (72% vs 67%)	Injection-site pain and upper respiratory tract infections	20 (18%)

MHD, migraine headache days; AEs, adverse events; NA, not available; sc, subcutaneous; iv, intravenous.

Table 3. TEV-48125 and AMG 334 for Migraine Prevention: Efficacy and Safety Results of Phase 2 Clinical Trials

Antibody	Study Population (Active vs Placebo)	Inclusion Criteria	Treatment Dose and Frequency (Patients)	Change of Primary Efficacy Outcome (Active vs Placebo)	AEs (Number of Patients, Active vs Placebo)	AEs	Patient Positive to Antidrug Antibodies
TEV-48125/ frestanezumab[58]	297 (193 vs 104)	High-frequency episodic migraine	225 mg sc, every 28 days (95)	−6.27 vs −3.46 ($P < .0001$)	44 vs 58 (46% vs 56%)	Injection-site pain and erythema, respiratory infections, and bronchitis	2 (1%)
			675 mg sc, every 28 days (96)	−6.09 vs −3.46 ($P < .0001$)	57 vs 58 (59% vs 56%)	Injection-site pain and erythema, sinusitis, and dizziness	
TEV-48125/ frestanezumab[59]	264 (175 vs 89)	Chronic migraine	675/225 mg sc, every 28 days (88)[a]	−59.84 vs −37.10 ($P = .0386$)	47 vs 36 (53% vs 40%)	Injection site-pain and pruritus, urinary tract infections, and sinusitis	2 (1%)
			900 mg sc, every 28 days (87)	−67.51 vs −37.10 ($P = .0057$)	41 vs 36 (47% vs 40%)	Injection site-pain and erythema, headache, and back-pain	
AMG334/ erenumab[60]	667	Chronic migraine	70 mg sc, every 28 days	−6.6 vs −4.2 ($P < .001$)	Not reported	Injection-site pain, upper respiratory tract infection, and nausea	NA
			140 mg sc, every 28 days	−6.6 vs −4.2 ($P < .001$)	Not reported		
AMG 334/erenumab[61]	483 (323 vs 160)	Episodic migraine	7 mg sc, every 28 days (108)	−2.2 vs −2.3	54 vs 82 (50% vs 54%)	Nasopharyngitis, fatigue, and headache	33 (10%)
			21 mg sc, every 28 days (108)	−2.4 vs −2.3	54 vs 82 (51 vs 54%)	Nasopharyngitis, back pain, and influenza	
			70 mg sc, every 28 days (107)	−3.4 vs −2.3 ($P = .021$)	57 vs 82 (54% vs 54%)	Nasopharyngitis, headache, nausea, and upper respiratory tract infections	

The primary efficacy outcome from baseline was MHD (or HH, in TEV–48125 utilized in chronic migraine) at 9–12 weeks.
MHD, migraine headache days; HH, headache hours; AEs, adverse events; NA, not available; sc, subcutaneously.
[a] In the first treatment cycle, 675 mg was administered; 225 mg was administered in the subsequent 2 treatment cycles.

administered intravenously to chronic migraineurs. This quarterly infusion formulation trial should be completed by November 2016 and has recruited 617 male and female patients between 18 and 55 years old. The first data collection was completed in March 2016 and showed (oral presentation at the 58th Annual Scientific Meeting of the American Headache Society in San Diego) that a single intravenous dose of eptinezumab at 30, 100, and/or 300 mg reduced migraine days to a similar extent for the entire 12-week period, whereas the 10-mg dose was subtherapeutic and similar to placebo. The most frequent adverse events observed were mild to moderate in severity (upper respiratory tract infection, dizziness, nausea, pharyngitis, sinusitis, and bronchitis); no drug-related safety signals and no infusion reactions occurred. The biopharmaceutical company has planned at least 2 phase 3 clinical trials for eptinezumab, including a double-blind, randomized, placebo-controlled trial (NCT02559895) to evaluate the efficacy and safety of 3 dose levels of eptinezumab administered intravenously as preventive treatment for frequent episodic migraineurs. The purpose is to recruit 600 patients of both sexes; they will be subdivided into 4 study arms (150 patients per group) to reach study completion in June 2017. In addition, a new double-blind, randomized, placebo-controlled, multidose trial named PROMISE2 is prepared to start. This study, the second pivotal trial of eptinezumab, plans to administer 2 dose levels of eptinezumab and placebo quarterly to chronic migraineurs and is likely to continue throughout 2018.

LY2951742/galcanezumab

LY2951742/galcanezumab is a fully humanized monoclonal antibody against CGRP developed by Eli Lilly and Company, with a half maximum inhibitory concentration of 30 pM. It was well tolerated in the first phase 1 studies, with a good safety profile as a single subcutaneous dose to 178 healthy males and females (NCT02576951), and as multiple doses given to 45 healthy Japanese and white participants (NCT02104765). Similar results were seen in 61 healthy white males receiving single (with doses ranging between 1 and 600 mg) or multiple (150 mg administered every 2 weeks for 6 weeks) subcutaneous injections (NCT01337596). It has a long elimination half-life (28 days), similar to ALD403, and the time to maximum serum concentration ranges from 7 to 13 days. The preliminary and encouraging results observed in phase 1 clinical trials have resulted in the development of at least 2 major phase 2 clinical trials. First, in a randomized, double-blind, placebo-controlled study, the efficacy and safety of LY2951742 were evaluated in the prevention of episodic migraine with or without aura (NCT01625988).[43] The study was conducted with 218 patients with 4 to 14 migraine headache days per month, randomly assigned (1:1) to LY2951742 (n = 108, but 1 patient withdrew before treatment) or placebo (n = 110). LY2951742 delivered subcutaneously (150 mg) once every 2 weeks for 12 weeks. The trial showed a significant reduction in the mean number of migraine headache days and a good tolerability profile. The post hoc efficacy analyses showed that 32% in the galcanezumab group versus 18% in the placebo group were complete responders. Several side effects occurred more frequently with galcanezumab with respect to placebo, such as injection-site pain and/or erythema (21 of 107 vs 7 of 110), upper respiratory tract infection (18 of 107 vs 10 of 110), and abdominal pain (6 of 107 vs 3 of 110). However, there were no serious adverse events related to the study drug. A second phase 2 study (NCT02163993) was conducted between August 2014 and October 2015 to assess the safety and efficacy of the compound in the prevention of episodic migraine. Four hundred ten male and female patients were recruited and were randomly assigned (2:1:1:1:1) to placebo (n = 137) or 1 of the 4 galcanezumab doses (n = 273). Subcutaneous injections of galcanezumab doses (5/50/120/300 mg) or placebo were delivered every 4 weeks during a 12-week treatment period. In accordance with the results of the previous phase 2a study, Eli Lilly and Company announced in a late-breaking session at the 57th Annual American Headache Society meeting in June 2015 that the compound had reached the primary end point in episodic migraine, proving a statistically significant reduction in migraine headache days and a good tolerability profile at all doses administered. Treatment related-adverse events that occurred in ≥5% of patients in any galcanezumab arm at rates greater than placebo included injection-site pain, upper respiratory tract infection, pharyngitis, and nausea. Because of the lack of further data, this clinical trial was not included in Table 2. Several phase 3 studies are in progress on galcanezumab. The first results will be announced starting in June 2017. The EVOLVE-1 (NCT02614183) and EVOLVE-2 (NCT02614196) studies are randomized, double-blind, placebo-controlled trials conducted with episodic migraineurs to evaluate the efficacy of subcutaneous galcanezumab (2 experimental doses) given once a month for 6 months. Each study will enroll 825 patients (EVOLVE-2 is currently recruiting participants) and will be completed in June 2017. The REGAIN study is another notable phase 3 clinical trial, carried out in chronic migraineurs in 106 study locations worldwide. The main purpose of this study (to be completed in April 2018) is to evaluate the efficacy of 2 subcutaneous experimental doses of galcanezumab in 825 patients, with monthly administration for 3 months. Finally, a long-term open-label safety study is in progress (NCT02614287) with 250 sufferers of migraine with or without aura, and some phase 3 studies on the efficacy, tolerability,

and long-term safety of the mAb are currently starting and ongoing, even including patients with episodic and/or chronic cluster headache (NCT02397473, NCT02438826, and NCT02797951). The first results will be announced starting in December 2016.

TEV-48125/frestanezumab

TEV-48125/frestanezumab, initially known as LBR-101, is a genetically engineered humanized mAb that has successfully completed a series of studies designed primarily to evaluate its safety and pharmacokinetic profile. The pooled results of the phase 1 program were published by Bigal and colleagues.[56] They reported that the compound was administered to 94 subjects, whereas the placebo was given to 45 people. TEV-48125 doses ranged from 0.2 to 2000 mg given either as a single intravenous infusion (day 1) or up to 300 mg given twice (day 1 and day 14); it was well tolerated at all doses. Treatment-related adverse events occurred in 21.2% of subjects receiving TEV-48125 (average = 1.4) compared with 17.7% of those receiving placebo (average = 1.3). Although the maximal tolerated dose has not been identified, the compound was not associated with any serious treatment-related adverse events and/or any clinically relevant patterns of changes in vital signs, electrocardiogram (ECG) parameters, or laboratory findings. The safety profile was further established by a subsequent double-blind, placebo-controlled study to assess the effects of sustained CGRP blockage on blood pressure and ECGs in healthy women (mean age, 56 years).[57] The 31 participants were randomly assigned to receive placebo or the mAb at doses up to 2000 mg. They were confined for 7 days and followed for 168 days. This study showed that sustained CGRP inhibition was not associated with hemodynamic or ECG changes in the study population. To summarize, the global results of the 6 phase 1 studies conducted to date were published in a recent review[42] that emphasized the excellent safety profile of the drug and discussed future drug development in phase 2 and phase 3 clinical trials. TEV-48125 was also recently investigated in 2 dedicated multicenter, randomized, double-blind, placebo-controlled phase 2b clinical trials with high-frequency episodic and chronic migraineurs. In one of these studies (NCT02025556), the efficacy and safety of 2 subcutaneous doses of TEV-48125 were assessed and compared with placebo for the preventive treatment of high-frequency episodic migraine (8–14 days per month) in 297 men and women. Between January and October 2014,[58] the participants were randomly assigned to receive placebo, 225 mg TEV-48125, or 675 mg TEV-48125 in three 28-day treatment cycles. Both doses of TEV-48125 reduced the least-squares mean (LSM) of migraine days ($P < .0001$) and the LSM of days with acute analgesic consumption compared with placebo (−3.10 days in the placebo group vs –4.86 days in the 225-mg dose group and –4.80 days in the 675-mg dose group). Treatment-related adverse events were reported by 24 patients in the placebo group (23%), 26 patients in the 225-mg dose group (27%), and 24 patients in the 675-mg group (25%); the majority of these events included mild injection-site pain or erythema. No serious treatment-related adverse events occurred. Only 1 severe treatment-related adverse event was recorded, and it was in the 225-mg dose group (severe injection-site pain). In a different phase 2b clinical trial (NCT02021773), it was evaluated whether monthly subcutaneous administration of TEV-48125 was safe and provided migraine prevention for chronic migraineurs.[59] Two hundred sixty-four patients were enrolled and randomly assigned to receive either placebo or 1 of the 2 doses of subcutaneous TEV-48125 in three 28-day treatment cycles. Compared with baseline, the mean change in number of headache-hours during weeks 9–12 was –59.84 hours (38%) in the 675/225-mg group and –67.51 hours (43%) in the 900-mg group, compared with –37.10 hours (22%) in the placebo group. The LSM difference was significant in both cases ($P = .0386$ and $P = .0057$, respectively). There were no qualitative or quantitative differences in treatment-related adverse events in the 3 cohorts of the study. The most frequent adverse events were minor injection-site reactions. No serious treatment-related adverse event occurred, and no relevant changes in blood pressure or other vital signs were recorded. Last, several phase 3 studies currently underway have the objective of assessing the efficacy and safety of subcutaneous administration of TEV-48125 in the preventive treatment of migraine (NCT02638103; NCT02629861; NCT02621931). The first results will not be available before October 2017.

AMG 334/erenumab

AMG 334/erenumab is a human IgG_2 that, unlike the previous mAbs, was developed to target and neutralize the CGRP receptor. Several phase 1 trials were performed to assess its safety, tolerability, and pharmacokinetic and pharmacodynamic properties. One of these trials (NCT01688739) was conducted with 48 healthy subjects and 20 migraineurs of both sexes, randomized into various cohorts, in which 6 dose levels of AMG 334 or placebo were administered as single subcutaneous or intravenous doses. In another similar phase 1 trial (NCT01723514), 40 subjects (24 healthy subjects and 16 migraineurs) were randomized into 5 subgroups. Each subject received 3 subcutaneous doses of either erenumab or placebo. These studies were completed in August 2013 and July 2014 with satisfactory results; the data have not been published. A further

phase 1 trial (NCT02741310), actually in progress, is considering the effect on blood pressure of erenumab given concomitantly with subcutaneous sumatriptan to 30 healthy subjects. August 2016 is the estimated date for final data collection and completion of the study. Last, an interesting phase 1 trial was developed to evaluate CGRP-receptor blockade by erenumab for preventing pituitary adenylate cyclase–activating polypeptide-38–induced migraine-like attacks in 42 migraineurs, with 1 to 5 migraine days per month. This study is actively recruiting participants and will end in February 2017. Erenumab was also implemented in several phase 2 clinical trials to evaluate its efficacy and safety for episodic and chronic migraine; some of the trials are still in progress. One of the first trials conducted (NCT02066415) was a randomized, double-blind, placebo-controlled study of chronic migraineurs. More than 650 subjects were randomly assigned to receive either placebo or 1 of the 2 AMG 334 subcutaneous doses every month for the 12-week treatment phase of this study. Significantly, more patients receiving the monthly mAb experienced a 50% or greater reduction in the number of monthly migraine days compared with placebo (40%, 41%, and 24%, respectively).[60] In addition, the safety profile of erenumab was similar to placebo across both treatment arms. No adverse events were reported in more than 5% of patients treated with the mAb. In a second trial (NCT01952574) the efficacy and safety of AMG 334 were evaluated in migraine prevention (with an inclusion criterion of 4 to 14 migraine days per month). In this randomized, double-blind, placebo-controlled study, 483 patients were randomly assigned to receive either monthly subcutaneous placebo or erenumab (7/21/70 mg) in a 3:2:2:2 ratio. The trial will be completed in November 2019; the first results were published recently.[61] The 70-mg dose was reported to significantly reduce the number of headache days, whereas the 7- and 21-mg doses showed no benefits compared with placebo. Only the patients receiving 70 mg reported greater reduction versus placebo in the number of days using acute medication (-2.5 vs -1.4, $P = .006$) and migraine-specific medication (-1.6 vs -0.7, $P = .004$). The safety profile of erenumab at all doses was similar to placebo; the number of migraineurs who had adverse events and the typology of the adverse events were similar among all treatment groups. No serious treatment-related adverse events were reported. The most common adverse events were nasopharyngitis, fatigue, headache, nausea, and back pain. An additional phase 2 clinical trial (NCT02174861) is under way to assess the long-term safety and efficacy of erenumab in chronic migraine prevention. It is a multicenter open-label study in which 612 patients of both sexes have received an open-label erenumab subcutaneous dose periodically for 13 months, followed by a safety follow-up visit. The study will be terminated in May 2017. To date, there are 2 phase 3 clinical trials on erenumab in progress, both of which are registered at the U.S. National Institutes of Health. One of these (NCT02483585), known as ARISE, is a randomized, double-blind, placebo-controlled study followed by an open-label treatment phase, with the aim of evaluating the effect of the compound in 577 episodic migraineurs randomly assigned 1:1 to placebo or erenumab. Preliminary results indicate that erenumab significantly reduced the mean number of headache days (–2.9) compared with placebo (–1.8) during the 12-week treatment phase. The second ongoing phase 3 trial (NCT02456740), conducted in 955 migraineurs with 1-year histories of episodic migraine randomly assigned to 1 of the 2 erenumab treatment groups or to placebo, has confirmed these findings. During the last 3 months of the double-blind treatment phase in this trial, patients experienced statistically significant reductions from baseline in monthly migraine days (–3.2 in the 70-mg group and –3.7 in the 140-mg group) compared with the placebo group (–1.8-day reduction). The clinical trial, called STRIVE, will end in June 2017.

Safety Concerns

The clinical use of mAbs was associated with a variety of immunological adverse events. They could be potential direct immunogens capable of generating hypersensitivity, autoimmunity, and acute infusion complications.[62] In the phase 2 clinical trials of prophylactic migraine therapy, the humanized anti-CGRP or anti-CGRP-receptor mAbs have not given rise to any serious immune-mediated adverse reactions, although they still contain nonhuman amino acids, presumably because the risk associated with the humanized monoclonal antibodies is very low. Although the first studies on these mAbs showed no relevant risk of immunological reactions, a small percentage of patients were positive for antidrug antibodies, in percentages ranging from 1% to 18%. The incidence of patients developing antidrug antibodies, the quantity generated, and their clinical significance are highly variable. Today it is impossible to make a prediction using preclinical safety immunological models; however, the implementation of novel, selected, and sensitive biomarkers could lead to early recognition of these specific antibodies in the clinical setting. This point is highly relevant because the antidrug antibodies may decrease therapy effectiveness and/or facilitate the manifestation of immunoallergic hypersensitivity reactions.[63,64] As an example, it is useful to repeat that in some chronic diseases, specifically rheumatoid arthritis[65] and multiple sclerosis,[66] the

emergence of antidrug antibodies was associated with reduced biological activity and consequent reduced therapeutic efficacy of the biological drug. In addition, the CGRP neuropeptide is involved in the regulation and homeostasis of many physiological processes, such as renal glomerular filtration,[67] bone metabolism,[68] gastric mucosal protection,[69] and several actions of the central nervous system.[70] It is particularly involved in cardiovascular system homeostasis, where it operates as a vasodilatory safeguard mechanism to prevent the complications of myocardial ischemia, such as myocardial infarction and heart failure,[71] hypertension,[72] and cerebral ischemia.[73] The neuropeptides represent the most potent endogenous vasodilator of coronary arteries,[74] and like nitrates, it relaxes them directly without mediation of endothelium-derived growth factors.[75] Little is known about the cardiovascular safety of CGRP system blockade with mAbs, and there is no evidence of treatment-related serious adverse cardiovascular events in phase 1 and phase 2 clinical trials. This is also true of the CGRP-receptor antagonists (gepants), which showed no particular cardiovascular safety issues in 2 prophylactic treatment trials.[39,52] Important research on the topic suggests cautious reassurance. Chronic treatment with anti-CGRP antibodies had no detectable effects on heart rate or blood pressure in monkeys and rats.[76,77] Moreover, CGRP antagonists seem to restore normal tonus in arteries dilated previously, but do not cause abnormal arterial constriction.[78] Regardless, it is our goal to understand whether blocking the CGRP system in migraineurs may be associated with an increased risk of cardiovascular events, especially in long-term use. This becomes even more relevant when one considers that migraine patients, particularly females, have an increased risk of stroke[79,80] and/or cardiovascular disease.[81,82] In the several studies conducted to date with anti-CGRP monoclonal antibodies, there was no evidence of other toxicity issues, and their overall safety profile is considered highly favorable. However, the adverse events because of chronic antagonism of CGRP biological activity need to be investigated in larger and extended clinical trials.

Patient-Focused Therapy

The numerous approved and unconventional treatments for migraine prophylaxis are frequently insufficient to control the painful attacks. Only a few patients are treatment responders, and about half of them have tolerability concerns or adverse reactions. Most of the drugs used for migraine prevention are anticonvulsants (such as topiramate and valproic acid), medicines with low therapeutic indexes and a prevalence of adverse drug reactions that varies between 10% and 40% when assessed by spontaneous reports or interviews.[83] The introduction of mAbs targeting the CGRP neuroactive peptide and/or its main receptor appears to lay the foundation for a new class of prophylactic drugs that could finally overcome, even only partially, the efficacy, safety, tolerability, and adherence issues that often affect chronic migraineurs. Regarding the available data directly comparing anti-CGRP mAbs with conventional migraine prevention treatments with high-quality evidence (topiramate and onabotulinumtoxinA), the monoclonal antibodies showed a significantly lower number of treatment-related adverse events, while maintaining effectiveness compared with placebo.[12,13,84] Their cost/benefit ratio represents one of the limiting factors to assess in the foreseeable future, as the uniqueness and complexity of monoclonal antibodies are publicized as reasons for the high expenses incurred by their users. They are so expensive in part because of the cost and difficulty of manufacturing. Furthermore, royalties and marketing costs are added to the total price, and the average cost for 1 year of treatment with one of the top 9 biologics in the United States reaches approximately $200 000.[85] Migraine is the most costly neurological disease for European society. The total annual cost of headache among European adults was estimated at €173 billion, and more than 80% of these expenditures were from migraine and its complications.[86] Accordingly, the high costs of mAbs should not affect health care policies, because increased investments in effective migraine care may reduce these losses and may be repaid severalfold by savings elsewhere,[87] in part by greater therapeutic adherence and fewer and milder adverse events related to this immunotherapy.

Conclusions

A migraine is one of the most disabling health problems worldwide, characterized by substantial disability in almost every aspect of life, including housework, employment, and social activities. Current pharmacotherapy options for migraine prophylaxis frequently show only modest efficacy, inconsistent responses to treatment, association with a high rate of adverse events, and poor tolerability. In these early clinical trials, anti-CGRP monoclonal antibodies have proven to be able to start another revolution in pharmacologic treatments for migraine, as triptans did a few decades ago, for acute migraine attacks. Despite the need for additional studies, especially in the long term, anti-CGRP mAbs have reduced the number of headache days and daily analgesic intake, with a safety profile similar to placebo. Today, monoclonal antibodies appear to offer a new and effective strategy for migraine prevention, representing more than hope for millions of chronic

migraineurs who are not currently taking any prophylactic therapy. We eagerly await the additional results of these novel medications.

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Declaration of Conflicting Interests

All the authors are actually involved in conducting the REGAIN study, a phase 3 clinical trial, to assess the long-term safety and tolerability of galcanezumab administered up to once monthly in chronic migraineurs. All the authors declare no other potential or ongoing conflicts of interest.

References

1. Stovner LJ, Zwart JA, Hagen K, Terwindt G, Pascual J. Epidemiology of headache in Europe. *Eur J Neurol*. 2006;13(4):333–345.
2. Stovner LJ, Andree C. Prevalence of headache in Europe: a review for the Eurolight project. *J Headache Pain*. 2010;11:289–299.
3. Steiner TJ, Stovner LJ, Vos T. GBD 2015: migraine is the third cause of disability in under 50s. *J Headache Pain*. 2016;17(1):104.
4. Buse DC, Manack AN, Fanning KM, et al. Chronic migraine prevalence, disability, and sociodemographic factors: results from the American Migraine Prevalence and Prevention Study. *Headache*. 2012;52:1456–1470.
5. Martelletti P. The therapeutic armamentarium in migraine is quite elderly. *Expert Opin Drug Metab Toxicol*. 2015;11(2):175–177.
6. Silberstein SD, Holland S, Freitag F. Evidence-based guideline update: pharmacologic treatment for episodic migraine prevention in adults. *Neurology*. 2012;78(17):1337–1345.
7. Wang SJ, Chen PK, Fuh JL. Comorbidities of migraine. *Front Neurol*. 2010;1:16.
8. Gracia-Naya M, Santos Lasaosa S, Rios-Gòmez C, et al. Predisposing factors affecting drop-out rates in preventive treatment in a series of patients with migraine. *Rev Neurol*. 2011;53:201–208.
9. Hepp Z, Dodick DW, Varon SF, Gillard P, Hansen RN, Devine EB. Adherence to oral-migraine-preventive medications among patients with chronic migraine. *Cephalalgia*. 2015;35:478–488.
10. Buse DC, Loder EW, Gorman JA, et al. Sex differences in prevalence, symptoms, and other features of migraine, probable migraine and other severe headache: results of the American Migraine Prevalence and Prevention Study. *Headache*. 2013;53:1278–1299.
11. Lipton RB, Bigal ME, Diamond M, et al. Migraine prevalence, disease burden, and the need for preventive therapy. *Neurology*. 2007;68(5):343–349.
12. Aurora SK, Dodick DW, Turkel CC, et al. OnabotulinumtoxinA for treatment of chronic migraine: results from the double-blind, randomized, placebo-controlled phase of the PREEMPT 1 trial. *Cephalalgia*. 2010;30:793–803.
13. Diener HC, Dodick DW, Aurora SK, et al. OnabotulinumtoxinA for treatment of chronic migraine: results from the double-blind, randomized, placebo-controlled phase of the PREEMPT 2 trial. *Cephalalgia*. 2010;30:804–814.
14. Negro A, Curto M, Lionetto L, Giamberardino MA, Martelletti P. Chronic migraine treatment: from OnabotulinumtoxinA onwards. *Expert Rev Neurother*. 2016;16(10):1217–1227.
15. Giamberardino MA, Martelletti P. Emerging drugs for migraine treatment. *Expert Opin Emerg Drugs*. 2015;20(1):137–147.
16. Edvinsson L. Functional role of perivascular peptides in the control of cerebral circulation. *Trends Neurosci*. 1985;8:126–131.
17. Edvinsson L. The journey to establish CGRP as a migraine target: a retrospective view. *Headache*. 2015;55:1249–1255.
18. Giamberardino MA, Affaitati G, Curto M, Negro A, Costantini R, Martelletti P. Anti-CGRP monoclonal antibodies in migraine: current perspectives. *Intern Emerg Med*. 2016;11(8):1045–1057.
19. Maggi CA. Tachykinins and calcitonin gene-related peptide (CGRP) as co-transmitters released from peripheral endings of sensory nerves. *Prog Neurobiol*. 1995;45:1–98.
20. Mulderry PK, Ghatei MA, Spokes RA. Differential expression of alpha-CGRP and beta CGRP by primary sensory neurons and enteric autonomic neurons on the rat. *Neuroscience*. 1988;25:195–205.
21. Russell FA, King R, Smillie S-J, Kodji X, Brain SD. Calcitonin gene-related peptide: physiology and pathophysiology. *Phys Rev*. 2014;94:1099–1142.
22. Brain SD, Williams TJ, Tippins JR, Morris HR, MacIntyre I. Calcitonin gene-related peptide is a potent vasodilator. *Nature*. 1985;313:54–56.
23. Edvinsson L, Fredholm BB, Hamel E, Jansen I, Verrecchia C. Perivascular peptides relax cerebral arteries concomitant with stimulation of cyclic adenosine monophosphate accumulation or release of an endothelium-derived relaxing factor in the cat. *Neurosci Lett*. 1985;58(2):213–217.
24. Oku R, Satoh M, Fujii N, Otaka A, Yajima H, Takagi H. Calcitonin gene-related peptide promotes mechanical nociception by potentiating release of substance P from

the spinal dorsal horn in rats. *Brain Res*. 1987;403:350–354.
25. Sun RQ, Tu YJ, Lawand NB, Yan JY, Lin Q, Willis WD. Calcitonin gene-related peptide receptor activation produces PKA- and PKC-dependent mechanical hyperalgesia and central sensitization. *J Neurophysiol*. 2004;92:2859–2866.
26. Uddman R, Edvinsson L, Ekman R, Kingman T, McCulloch J. Innervation of the feline cerebral vasculature by nerve fibers containing calcitonin gene-related peptide: trigeminal origin and co-existence with substance P. *Neurosci Lett*. 1985;62(1):131–136.
27. Bahra A, Matharu MS, Buchel C, Frackowiak RS, Goadsby PJ. Brainstem activation specific to migraine headache. *Lancet*. 2001;357:1016–1017.
28. Afridi SK, Matharu MS, Lee L, et al. A PET study exploring the laterality of brainstem activation in migraine using glyceryl trinitrate. *Brain*. 2005;128:932–939.
29. Moskowitz MA, Nozaki K, Kraig RP. Neocortical spreading depression provokes the expression of c-fos protein-like immunoreactivity within trigeminal nucleus caudalis via trigeminovascular mechanisms. *J Neurosci*. 1993;13:1167–1177.
30. Goadsby PJ. Recent advances in understanding migraine mechanisms, molecules and therapeutics. *Trends Mol Med*. 2007;13:39–44.
31. Edvinsson L, Goadsby PJ. Neuropeptides in migraine and cluster headache. *Cephalalgia*. 1994;14:320–327.
32. Lassen LH, Haderslev PA, Jacobsen VB, Iversen HK, Sperling B, Olesen J. CGRP may play a causative role in migraine. *Cephalalgia*. 2002;22(1):54–61.
33. Stepień A, Jagustyn P, Trafny EA, Widerkiewicz K. Suppressing effect of the serotonin 5HT1B/D receptor agonist rizatriptan on calcitonin gene-related peptide (CGRP) concentration in migraine attacks. *Neurol Neurochir Pol*. 2003;37(5):1013–1023.
34. Cernuda-Morollón E, Larrosa D, Ramón C, Vega J, Martínez-Camblor P, Pascual J. Interictal increase of CGRP levels in peripheral blood as a biomarker for chronic migraine. *Neurology*. 2013;81(14):1191–1196.
35. Diener HC, Barbanti P, Dahlöf C, Reuter U, Habeck J, Podhorna J. BI 44370 TA, an oral CGRP antagonist for the treatment of acute migraine attacks: results from a phase II study. *Cephalalgia*. 2011;31(5):573–584.
36. Marcus R, Goadsby PJ, Dodick D, Stock D, Manos G, Fischer TZ. BMS-927711 for the acute treatment of migraine: a double-blind, randomized, placebo controlled, dose-ranging trial. *Cephalalgia*. 2014;34(2):114–125.
37. Olesen J, Diener HC, Husstedt IW, et al. Calcitonin gene-related peptide receptor antagonist BIBN 4096 BS for the acute treatment of migraine. *N Engl J Med*. 2004;350(11):1104–1110.
38. Cui XP, Ye JX, Lin H, Mu JS, Lin M. Efficacy, safety, and tolerability of telcagepant in the treatment of acute migraine: a meta-analysis. *Pain Pract*. 2015;15(2):124–131.
39. Ho TW, Connor KM, Zhang Y, et al. Randomized controlled trial of the CGRP receptor antagonist telcagepant for migraine prevention. *Neurology*. 2014;83(11):958–966.
40. Edvinsson L. CGRP receptor antagonists and antibodies against CGRP and its receptor in migraine treatment. *Br J Clin Pharmacol*. 2015;80(2):193–199.
41. Dodick DW, Goadsby PJ, Silberstein SD, et al. Safety and efficacy of ALD403, an antibody to calcitonin gene-related peptide, for the prevention of frequent episodic migraine: a randomised, double-blind, placebo-controlled, exploratory phase 2 trial. *Lancet Neurol*. 2014;13:1100–1107.
42. Walter S, Bigal ME. TEV-48125: a review of a monoclonal CGRP antibody in development for the preventive treatment of migraine. *Curr Pain Headache Rep*. 2015;19:6.
43. Dodick DW, Goadsby PJ, Spierings EL, et al. Safety and efficacy of LY2951742, a monoclonal antibody to calcitonin gene-related peptide, for the prevention of migraine: a phase 2, randomized, double-blind, placebo-controlled study. *Lancet Neurol*. 2014;13:885–892.
44. Shi L, Lehto SG, Zhu DX, et al. Pharmacologic characterization of AMG334, a potent and selective human monoclonal antibody against the calcitonin gene-related peptide receptor. *J Pharmacol Exp Ther*. 2016;356:223–231.
45. Wrobel Goldberg S, Silberstein SD. Targeting CGRP: a new era for migraine treatment. *CNS Drugs*. 2015;29:443–452.
46. Yu YJ, Watts RJ. Developing therapeutic antibodies for neurodegenerative disease. *Neurotherapeutics*. 2013;10:459–472.
47. Lundblad C, Haanes KA, Grände G, Edvinsson L. Experimental inflammation following dural application of complete Freund's adjuvant or inflammatory soup does not alter brain and trigeminal microvascular passage. *J Headache Pain*. 2015;16:91.
48. Eftekhari S, Salvatore CA, Johansson S, Chen T, Zeng Z, Edvinsson L. Localization of CGRP, CGRP receptor, PACAP and glutamate in rhesus monkey trigeminal ganglion. Relation to the blood-brain barrier. *Brain Res*. 2015;1600:93–109.
49. Schankin CJ, Maniyar FH, Seo Y, et al. Ictal lack of binding to brain parenchyma suggests integrity of the blood-brain barrier for 11C-dihydroergotamine during glyceryl trinitrate-induced migraine. *Brain*. 2016;139:1994–2001.
50. Hansel TT, Kropshofer H, Singer T, Mitchell JA, George AJ. The safety and side effects of monoclonal antibodies. *Nat Rev Drug Discov*. 2010;9(4):325–338.
51. Zhou H, Mascelli MA. Mechanisms of monoclonal antibody-drug interactions. *Annu Rev Pharmacol Toxicol*. 2011;51:359–372.

52. Ho TW, Ho AP, Ge YJ, et al. Randomized controlled trial of the CGRP receptor antagonist telcagepant for prevention of headache in women with perimenstrual migraine. *Cephalalgia*. 2016;36(2):148–161.
53. Karasek C, Ojala E, Allison D, Latham J. Characterization of the intrinsic binding features of three anti-CGRP therapeutic antibodies effective in preventing migraine: a comparative pre-clinical case study of ALD403, LY2951742, TEV48125. 58th Annual Scientific Meeting of the American Headache Society; June 2016, San Diego, California.
54. Baker B, Hodsman P, Smith J. PK & PD supporting a single dose, placebo-controlled randomized ascending dose study of ALD403, a humanized anti-calcitonin gene-related peptide (CGRP) monoclonal antibody administered IV or SC. 17th Congress of the International Headache Society; May 2015, Valencia, Spain.
55. Baker B, Schaeffler B, Pederson S, Potter T, Smith J. A multiple-dose, placebo-controlled, randomized phase I clinical trial of ALD403, an anti-calcitonin gene-related peptide monoclonal antibody, administered once every 3-months via IV, SC, or IM. 58th Annual Scientific Meeting of the American Headache Society; June 2016, San Diego, California.
56. Bigal ME, Escandon R, Bronson M, et al. Safety and tolerability of LBR-101, a humanized monoclonal antibody that blocks the binding of CGRP to its receptor: results of the Phase 1 program. *Cephalalgia*. 2013;34: 483–492.
57. Bigal ME, Walter S, Bronson M, Alibhoy A, Sager P, Escandon R. Cardiovascular and hemodynamic parameters in women following prolonged CGRP inhibition using LBR-101, a monoclonal antibody against CGRP. *Cephalalgia*. 2014;34:968–976.
58. Bigal ME, Dodick DW, Rapoport AM, et al. Safety, tolerability and efficacy of TEV-48125 for preventive treatment of high-frequency episodic migraine: a multicentre, randomised, double-blind, placebo-controlled, phase 2b study. *Lancet Neurol*. 2015;14:1081–1090.
59. Bigal ME, Edvinsson L, Rapoport AM, et al. Safety, tolerability, and efficacy of TEV-48125 for preventive treatment of chronic migraine: a multicentre, randomised, double-blind, placebo-controlled, phase 2b study. *Lancet Neurol*. 2015;14:1091–1100.
60. Tepper S, et al. Phase II, randomized, double-blind, placebo-controlled study to evaluate the efficacy and safety of erenumab (AMG 334) in chronic migraine prevention. 5th European Headache and Migraine Trust Annual Congress; September 2016, Glasgow, UK.
61. Sun H, Dodick DW, Silberstein S, et al. Safety and efficacy of AMG 334 for prevention of episodic migraine: a randomised, double-blind, placebo-controlled, phase 2 trial. *Lancet Neurol*. 2016;15(4):382–390.
62. Descotes J. Immunotoxicity of monoclonal antibodies. *MAbs*. 2009;1(2):104–111.
63. Stallmach A, Giese T, Schmidt C, Meuer SC, Zeuzem SS. Severe anaphylactic reaction to infliximab: successful treatment with adalimumab – report of a case. *Eur J Gastroenterol Hepatol*. 2004;16:627–630.
64. Pharand C, Palisaitis DA, Hamel D. Potential anaphylactic shock with abciximab readministration. *Pharmacotherapy*. 2002;22:380–383.
65. Mok CC, van der Kleji D, Wolbink GJ. Drug levels, anti-drug antibodies, and clinical efficacy of the anti-TNFα biologics in rheumatic diseases. *Clin Rheumatol*. 2013;32(10):1429–1435.
66. Bertolotto A. Evaluation of the impact of neutralizing antibodies on IFNβ response. *Clin Chim Acta*. 2015;449:31–36.
67. Gnaedinger MP, Uehlinger DE, Weidmann P, et al. Distinct hemodynamic and renal effects of calcitonin gene-related peptide and calcitonin in man. *Am J Physiol*. 1989;257:E848–E854.
68. Bernard GW, Shih C. The osteogenic stimulating effect of neuroactive calcitonin gene-related peptide. *Peptides*. 1990;11:625–632.
69. Lenz HJ, Mortrud MT, Rivier JE, Brown MR. Central nervous system actions of calcitonin gene-related peptide on gastric acid secretion in the rat. *Gatroenterology*. 1985;88:539–544.
70. Fischer JA, Born W. Calcitonin gene products: evolution, expression and biological targets. *Bone Miner*. 1987;2:347–359.
71. Li J, Levick SP, Dipette DJ, Janicki JS, Supowit SC. Alpha-calcitonin gene-related peptide is protective against pressure overload-induced heart failure. *Regul Pept*. 2013;185:20–28.
72. Smillie SJ, King R, Kodji X, et al. An ongoing role of alpha-calcitonin gehe-related peptide as part of a protective network against hypertension, vascular hypertrophy, and oxidative stress. *Hypertension*. 2014;63: 1056–1062.
73. Zhang F, Wang S, Signore AP, Chen J. Neuroprotective effects of leptine against ischemic injury induced by oxigen-glucose deprivation and transient cerebral ischemia. *Stroke*. 2007;38:2329–2336.
74. Preibisz JJ. CGRP and regulation of human cardiovascular homeostasis. *Am J Hypertens*. 1993;6:434–450.
75. Foulkes R, Shaw N, Hughes B. The vasodilatory effects of repeated CGRP and nitrate administration in small pig coronary arteries. *Regul Pept*. 1989;25:25–36.
76. Walter S, Alibhoy A, Escandon R, Bigal ME. Evaluation of cardiovascular parameters in cinomolgus monkeys following IV administration of LBR-101, a monoclonal antibody against calcitonin gene-related peptide. *MAbs*. 2014;6:871–878.
77. Zeller J, Poulsen KT, Sutton JE, et al. CGRP function-blocking antibodies inhibit neurogenic vasodilatation without affecting heart rate or arterial blood pressure in the rat. *Br J Pharmacol*. 2008;155:1093–1103.

78. Verheggen R, Bumann K, Kaumann AJ. BIBN4096BS is a potent competitive antagonist of the relaxant effects of alpha-CGRP on human temporal artery: comparison with CGRP(8-37). *Br J Pharmacol*. 2002;136(1):120–126.
79. Schürks M, Rist PM, Bigal ME, Buring JE, Lipton RB, Kurth T. Migraine and cardiovascular disease: systematic review and meta-analysis. *BMJ*. 2009;339: b3914.
80. Spector JT, Kahn SR, Jones MR, Jayakumar M, Dalal D, Nazarian S. Migraine headache and ischemic stroke risk: an updated meta-analysis. *Am J Med*. 2010;123:612–624.
81. Bigal ME, Kurth T, Santanello N, et al. Migraine and cardiovascular disease: a population-based study. *Neurology*. 2010;74:628–635.
82. Kurth T, Schürks M, Logroscino G, Gaziano JM, Buring JE. Migraine, vascular risk, and cardiovascular events in women: prospective cohort study. *BMJ*. 2008;337:a636.
83. Perucca P, Gilliam FG. Adverse effects of antiepileptic drugs. *Lancet Neurol*. 2012;11(9):792–802.
84. Silberstein SD, Lipton RB, Dodick DW, et al. Efficacy and safety of topiramate for the treatment of chronic migraine: a randomized, double-blind, placebo-controlled trial. *Headache*. 2007;47:170–180.
85. Shaughnessy AF. Monoclonal antibodies: magic bullets with a hefty price tag. *BMJ*. 2012;345:e8346.
86. Linde M, Gustavsson A, Stovner LJ, et al. The cost of headache disorders in Europe: the Eurolight project. *Eur J Neurol*. 2012;19:703–e43.
87. Steiner TJ. Headache burdens and bearers. *Funct Neurol*. 2000;15(Suppl 3):219–223.