



## Review Article

## Preclinical perspectives on disorders of the temporomandibular joint: Tracing the past, navigating the present, and shaping the future

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## ABSTRACT

Temporomandibular disorders (TMDs) are complex conditions characterized by orofacial pain and dysfunction, affecting a significant portion of the population. TMDs may involve joint and/or muscle pain, dysfunction (e.g., noise, limited or altered jaw movements), or both, leading to a marked decrease in quality of life. Patients often experience functional limitations that hinder eating, speaking, and daily activities. Additionally, TMDs are frequently associated with psychological distress, including anxiety and depression, which further impacts overall well-being. Despite the profound individual and societal impact of TMDs, effective therapies remain

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elusive, partly due to deficiencies in translational research. A primary limitation in the TMD field is the scarcity of animal models that accurately replicate disease features in humans. This may ultimately be due to species differences, but likely also reflects the etiological and symptomatic heterogeneities of TMDs, as there are over 30 different conditions in this umbrella term. Both factors pose a significant challenge in developing and using animal models for TMD research. This review highlights preclinical TMD research to enhance clinical care, focusing on anatomy/physiology, pain and behavior models, functional and tissue modeling, biopsychosocial factors, and technological considerations. The "TMD Research Community" collaborated to produce this review, with the Discussion offering a proposal for a path forward.

**Perspective**

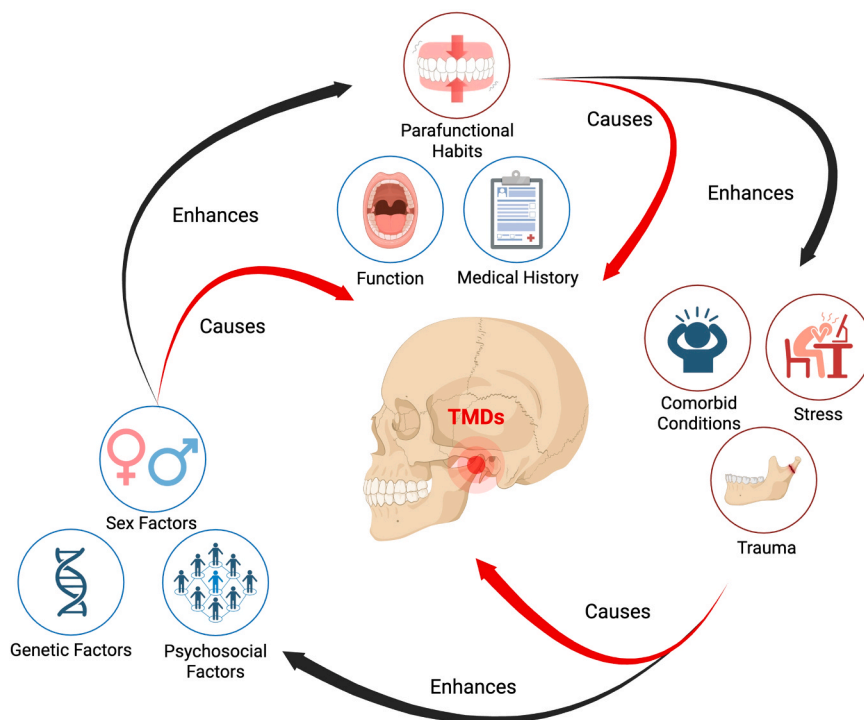
This review highlights the current landscape of preclinical TMD research in relation to translational research and clinical therapy development. It outlines collaborative strategies for future studies as a roadmap to improve success in TMD research and patient care.

**Introduction - clinical to preclinical reverse translation**

Temporomandibular disorders (TMDs) are a group of over 30 complex clinical conditions characterized by orofacial pain and dysfunction, often overlapping with other chronic pain conditions (COPCs). Painful TMDs affect approximately 5% of the adult population, producing substantial societal costs and personal suffering.<sup>1</sup> TMDs are more prevalent and more severe among women than men<sup>1</sup> and have an earlier age of onset than most musculoskeletal diseases of appendicular joints. While often viewed and treated as localized dental disorders, most TMDs are complex systemic medical conditions driven by a mosaic of biopsychosocial factors often superimposed on local musculoskeletal dysfunction. Indeed, the Orofacial Pain: Prospective Evaluation and Risk Assessment (OPPERA) studies identified multiple clinical, psychosocial, and biological factors associated with chronic TMDs as well as factors predicting risk for first onset TMD.<sup>2-5</sup> Moreover, up to 80% of individuals with a painful TMD also report experiencing one or more additional chronic pain conditions.<sup>6,7</sup> Like many other chronic pain conditions, TMDs arise from a complex interplay of biological,

psychological, and social factors, including genetic predisposition, sex-related differences, and stress (see Fig. 1). Distinctive to TMDs are orofunctional contributors such as masticatory biomechanics, parafunctional behaviors (e.g., bruxism, clenching), and unique anatomical features. Notably, the mandible functions as a rigid bridge between the bilateral TMJs, limiting joint rest due to continuous functional demands like chewing and speaking—an anatomical constraint not typically observed in other chronic pain disorders.

Although the OPPERA studies have provided valuable insights, the etiopathogenesis of TMDs remains incompletely understood. This knowledge gap is largely attributable to the complexity of elucidating the underlying mechanisms that drive the initiation and chronicity of TMDs. Compounding this challenge is the limited availability of preclinical models that are both mechanistically informative and clinically translatable.<sup>8,9</sup> Nevertheless, the development of precision diagnostics and targeted therapeutics necessitates the use of well-validated preclinical models to interrogate both disease mechanisms and therapeutic efficacy. In the context of translational research, it is essential to integrate knowledge of current clinical management strategies (see Table 1) with the development of novel interventions. A critical consideration in the use of animal models is that they often reflect more invasive or high-risk treatment paradigms, whereas clinical care typically begins with conservative approaches and escalates as needed. This contrast highlights the essential function of preclinical models in bridging mechanistic research with the development of clinically meaningful and patient-focused therapies.



**Fig. 1.** Multifactorial mechanisms underlying the development and exacerbation of TMDs. Created in BioRender. Neubert, J. (2025) <https://BioRender.com/s4bqg83>

As with many types of chronic pain disorders, one limitation in the TMD field has been the lack of animal models that can recapitulate all disease features in humans for any one TMD condition. This may ultimately prove to be due to species differences, but is also likely a reflection of the etiological and symptomatic heterogeneities of individual TMD conditions and their overlap, which can result from various factors, including biomechanical, inflammatory, neurovascular, psychosocial, and genetic predispositions.<sup>4,10-12</sup> Trauma to the jaw or face, parafunctional behaviors, systemic conditions such as arthritis, hormonal fluctuations, and psychological stressors are among the multifactorial causes and/or enhancers implicated in the development and maintenance of TMDs (Fig. 1). Symptomatically, TMDs can present with a wide array of signs and symptoms, including joint and/or muscle pain, restricted mandibular movement, clicking or popping sounds in the joint, muscle tenderness, headache, ear pain, neck pain, sleep bruxism, and psychological comorbidities.<sup>4,13,14</sup> Furthermore, the unique epidemiology of TMDs, and the distinct anatomy, function, development, histology and cellularity of the temporomandibular joint (TMJ) suggest that findings from other disorders of musculoskeletal structures cannot be applied readily to TMDs. Research on TMDs shows that biopsychosocial factors play a significant role in the onset and persistence of severe, long-term pain. Therefore, it is essential for preclinical TMD models (targeting at least one TMD condition) to consider not just the biological factors, but the psychosocial factors as far as possible, despite the limitations of animal studies. And while the psychological and social influences on pain have been largely overlooked in preclinical studies, as described below, there is a growing appreciation of the impact of these variables on the trajectory of a TMD model, as well as the identification of strategies to assess the impact of these variables on critical endpoints in preclinical studies. This review highlights the current state of preclinical TMDs research and addresses questions of how best to move the field forward to maximize clinical translatability and ultimately better patient care for those suffering from a TMD.

## Anatomy, physiology, and nociceptive pathways of the TMJ and masticatory muscles

*This section highlights the anatomical and physiological components contributing to TMDs in humans and examines the similarities and differences in preclinical animal models.*

TMDs are a leading cause of nonodontogenic orofacial pain that includes multiple clinical conditions caused by the impairment of structural or functional properties of masticatory muscles. The most frequently described symptoms are joint and muscle pain and dysfunction. While TMDs are a multifactorial set of conditions, the development and progression of a TMD may be related to several different factors. As such, it is important to understand the anatomical features involved in TMDs including the myogenic and arthrogenic components, as both can differ in etiology, pathophysiology, and clinical signs and symptoms.

### TMJ – normal anatomy and function

The TMJ, which is also referred to as the mandibular joint or jaw joint in humans, is defined as a bicondylar articulation that acts like a sliding hinge between the lower jaw and the skull. The TMJ components include the temporal and mandibular bones, the intra-articular disc, fibrous capsule, synovial membranes and fluid, and supporting ligaments.<sup>15</sup> A key structure is the articular disc, which has an elliptical shape that is responsible for absorbing and distributing the masticatory force, minimizing bone instability, thus contributing to the normal movement of the joint.<sup>16</sup> The articular disc divides the TMJ into two compartments that are filled with synovial fluid. The upper compartment, which experiences translational movement of the joint,<sup>17</sup> is formed by the glenoid fossa of the temporal bone. The lower compartment experiences rotational movements between the articular disc and the mandibular condyle.<sup>16</sup>

The synovial membrane of the TMJ is well-supplied with blood vessels and nerves. The synovial fluid, which is rich in hyaluronan and lubricin, provides essential nutrients and immune support to the intra-

**Table 1**  
Existing and emerging treatments for TMD.

Non-surgical Treatments
<u>Medications</u>
NSAIDs – <i>ibuprofen</i>
Muscle relaxants – <i>baclofen, flexeril</i>
Tricyclic antidepressants – <i>nortriptyline, amitriptyline</i>
<u>Physical Therapy</u>
Modalities – <i>heat/ice, ultrasound, TENS</i>
Exercises – <i>passive jaw and neck muscle stretching</i>
Massage and posture training
<u>Behavioral Therapy</u>
Relaxation and stress management - <i>deep breathing, meditation, and mindfulness</i>
Cognitive behavioral therapy/counseling – <i>self-care to avoid harmful habits</i>
Reduction of negative operant behaviors
<u>Other Conservative Treatments</u>
Occlusal appliances - <i>balanced hard flat plane splints</i>
Dietary changes - <i>soft food diets</i>
Trigger point injections – <i>dry needling</i>
Acupuncture
Joint Capsule Injections
Hyaluronic acid
Corticosteroids
Surgical Treatments
Arthrocentesis – <i>joint lavage</i>
Arthroscopy - <i>minimally invasive procedure</i>
Arthroplasty - <i>reconstruction of the joint</i>
Joint replacement
Emerging Treatments
Botox – <i>onabotulinumtoxinA</i>
Biologics - <i>CGRP monoclonal antibodies</i>
Cannabinoids – <i>CBD (medical marijuana)</i>

articular tissues.<sup>18–21</sup> In fact, synovial fluid serves a dual purpose: it nourishes the avascular articular surfaces and acts as a lubricant, thereby reducing friction during joint movement.<sup>22</sup>

**Innervation** of the TMJ in humans and rodents comprises various nerve fiber types responsible for sensory reception, motor function, autonomic vascular regulation, and immune modulation. The trigeminal innervation of the TMJ primarily involves the auriculotemporal nerve posteriorly, the masseteric nerve anteriorly, the posterior deep temporal nerve anteromedially, and a TMJ branch from the mandibular nerve medially.<sup>23</sup> The vast majority of what is known about the innervation of the TMJ has been gleaned from studies of preclinical models, largely rats and mice. These data suggest that sensory innervation of the joint includes nociceptive fibers expressing transient receptor potential vanilloid 1 (TRPV1) (approximately 50% of trigeminal fibers in the TMJ (unpublished findings)<sup>24</sup>), Calcitonin Gene-Related Peptide (CGRP) and substance P,<sup>25</sup> and P2X receptors.<sup>26,27</sup> In addition, nerve fibers exhibiting neuropeptide Y (NPY) or tyrosine hydroxylase (TH, sympathetic fibers) are assumed to be found in the TMJ capsule, disc attachment, the peripheral portion of the articular disc, periosteum, synovial membrane, and surrounding muscles. No fibers are seen in the central portion of the disc. The distribution of TH-immunoreactive fibers is almost the same as that of NPY-immunoreactive nerves. Most of these nerves appear to be closely associated with the vasculature and are lost following lesioning of the stellate sympathetic ganglia.<sup>28</sup> Retrograde tracers injected in the rodent joint cavity label the sensory trigeminal ganglion and cervical dorsal root ganglia extending to the fifth vertebrae. The superior cervical and stellate sympathetic ganglia are also labeled.<sup>28,29</sup> Parasympathetic innervation of the joint is supplied by sphenopalatine, nodose, and otic ganglia.<sup>30,31</sup> The rat masseter and temporalis muscles are innervated by fast conducting (A-alpha, A-beta) fibers from the mesencephalic nucleus in the brain stem, and by slower conducting (A-delta and C) fibers from the trigeminal ganglion.<sup>32,33</sup> With a growing body of evidence suggesting that there are species differences in afferent properties, in particular, the overlap in putative nociceptive signaling molecules and receptors,<sup>34</sup> much of this data will need to be validated in human tissue.

TMDs frequently involve a **masticatory muscle component**. These muscles are responsible for ensuring the appropriate function of the joint. They are fixed to the cranium and connect to the jaw.<sup>35</sup> They are divided into abductors (temporalis, masseter, and medial pterygoid muscles) and adductors (lateral pterygoid and submandibular muscles). The lateral pterygoid muscles are involved in jaw protrusion and help with lateral movements.<sup>35</sup> The masseteric nerve innervates the masseter muscle, while the posterior deep temporal nerve supplies the temporal muscle.<sup>23</sup> Masticatory muscle afferent fibers with mechanical activation thresholds considered nociceptive are predominantly A-delta fibers.<sup>32</sup> These putative nociceptors also respond to injection of noxious substances, such as hypertonic saline, potassium chloride, glutamate, and serotonin (5-HT), into their muscle mechanoreceptive fields. This suggests that many are polymodal nociceptors.<sup>32,36,37</sup> Afferent discharge evoked by glutamate and serotonin is mediated by activation of N-methyl-D-aspartate (NMDA) and 5-HT<sub>3</sub> receptors, respectively, which are expressed on a subgroup of masticatory muscle afferent fibers. The magnitude of masticatory muscle afferent discharge evoked by activation of NMDA receptors is greater in female than in male rats.<sup>38</sup> Masticatory muscle afferent fibers also express estrogen receptors<sup>39</sup> and the magnitude of NMDA-evoked afferent discharge is positively correlated with blood estrogen levels in female rats.<sup>38,39</sup> There is a strong central projection of masticatory muscle nociceptive fibers from the trigeminal ganglion to the trigeminal subnucleus caudalis, where nociceptive second order neurons are located.<sup>40</sup>

A stable dentition can help in providing an even distribution of forces across the TMJ and masticatory muscles. Given that, an increasing number of preclinical studies use models involving induction of a change in load distribution with occlusal interference and occlusal adjustments to investigate mechanisms, pathology, and potential treatments for TMD. For example, a change in occlusion enhances glycolytic

metabolism, alters vascularization, and increases oxidative stress in the medial pterygoid muscle.<sup>41,42</sup> Not surprisingly, the change in occlusion-associated effects is more pronounced when combined with acute stress, which is a significant risk factor for human TMD. Abdalla et al. reported that alterations in a mechanical loading model led to significantly elevated levels of pro-inflammatory cytokines such as tumor necrosis factor-alpha (TNF- $\alpha$ ), interleukin (IL)-1 $\beta$ , IL-6, fractalkine (CX3CL1), and TNF- $\alpha$  converting enzyme (ADAM-17).<sup>43</sup>

This was accompanied by increased satellite glial cell activation and elevated release of substance P and glutamate in the trigeminal ganglion. Of note, these change in occlusion models are meant to recapitulate TMJ articular surface damage and muscle pain, and do not model the etiology of any known TMDs. The closest scenarios would be condyle fracture from trauma or orthognathic surgery.

#### *Human versus non-human TMJ – key similarities and differences*

The TMJ is the last joint to appear in mammalian evolution and a defining feature of mammals. The TMJ serves a critical function in humans and other mammals, exhibiting remarkable **similarities** in its anatomical and functional characteristics.<sup>17,44–49</sup> The fundamental components of the TMJ such as the mandibular condyle, temporal bone, articular disc, synovial membrane, and synovial fluid, are conserved across mammalian species. The presence of an articular disc, which acts as a cushioning structure between the bones, is critical for efficient joint movement. Humans and other mammals share a similar composition of this disc, particularly its collagen and elastin fibers, providing comparable mechanical properties.<sup>44,46,47</sup> Functionally, the TMJ facilitates rotational and/or translational movements essential for mastication and other oral functions. The mechanics of these movements have been observed to be quite similar across species.<sup>44</sup> For example, the forces generated during biting and chewing are analogous in humans, dogs, and primates, which suggests a shared evolutionary adaptation to dietary needs. In addition, human and animal TMJs contain similar cell types, such as osteocytes, osteoclasts, chondrocytes, synoviocytes, fibroblasts, and adipocytes.<sup>50–53</sup>

There are also notable **differences** between animal and human TMJs.<sup>45,46,48,54</sup> Rodent (rat, mice) TMJs differ from human TMJs in several aspects: in rodents the glenoid fossa is shallow and flat, the lateral pterygoid muscle is less voluminous and has less functional force, and the upper articular cavity forms first, followed by the lower articular cavity. The articular disc rarely becomes fibrous cartilage with aging in rats as is observed in the human TMJ.<sup>55</sup> Although the condyles are covered by a thick layer of hyaline cartilage in both human and rat TMJs, human condyles typically have more layers of chondrocytes.<sup>56</sup> The human TMJ is much larger than that of rodents. A key distinction is the absence of an articular eminence in rodents, which allows for extensive protrusive movements.<sup>45,56,57</sup> Human TMJs are highly specialized joints that consist of a biconcave fibrocartilaginous disc positioned between the mandibular condyle and the articular fossa of the temporal bone. This unique arrangement allows for smooth and coordinated jaw movement during activities such as chewing, speaking, and yawning. In contrast, many animal TMJs lack this intricate disc-condyle structure. For example, rodents have simpler TMJ anatomy, with a more rudimentary disc and a less developed articular fossa. Furthermore, rodents have a different masticatory system, primarily using their incisors for gnawing, whereas humans primarily use their molars for chewing.<sup>58</sup> This difference in chewing patterns can influence the way TMD-like symptoms manifest in animal models and may impact the applicability of research findings to human TMD. Another major difference is that carnivore species have a TMJ that only rotates and cannot dislocate, and herbivore species have mainly translatory movements, all with differences in the size ratio of the masseter to the temporalis muscles when compared to omnivores (humans). Functional differences in pain processing also pose challenges for TMD research. Humans have a complex and subjective experience of pain, influenced

by psychological, social, and emotional factors, which may not be fully replicated in animal models.<sup>59,60</sup>

## Rodent models of TMDs (Fig. 2)

*This section highlights small animal (mostly rodent) models of TMD*

The main goal of any disease model is ultimately to develop more effective therapeutic strategies. These are generally believed to arise through a better mechanistic understanding of the disease, if not the ability to establish causal links between putative mechanisms and disease phenotypes via experiments that would not be possible in patients. This has been the case in the development of preclinical models of TMDs. Furthermore, because pain is one of the most debilitating symptoms of TMDs, a primary focus of TMD models has been to recapitulate what is interpreted as pain in the TMJ and musculature complex as reported in humans.

Animal models of TMDs have been induced through chemical, mechanical, surgical, or genetic approaches,<sup>8,9,44,55,61–63</sup> which have more recently been combined with the manipulation of other environmental factors such as social interactions, stress and diet, given the growing appreciation of the complex array of factors that influence the manifestation of TMDs in patients. The heterogeneity of TMDs poses a particular challenge for developing animal models in TMD research, where any single model is often dismissed for failing to capture the heterogeneity observed in the patient population. Additionally, incorporating multiple risk factors of TMD pain etiology from any one TMD condition into a single model is difficult. This limitation can affect the translational relevance of findings from animal studies to humans. Animal models of TMDs have been comprehensively reviewed elsewhere.<sup>8,9,44,55,61–63</sup> To avoid redundancy with existing literature, we will describe these models, focusing on strengths and weaknesses.

**Chemical models.** Though TMDs have multifactorial etiologies,<sup>10–12,64–66</sup> inflammation of the TMJ and/or masticatory muscles contribute to TMD development in subgroups of patients. Directly injecting chemical agents into the TMJ or surrounding musculature is relatively easy and reproducible and generates an inflammatory response with the intention of reproducing this aspect of the clinical phenotype. The choice of chemical is largely driven by the endpoint, where acute nociceptive behavior is generated by the injection of chemicals like capsaicin or formalin, while a more slowly developing but robust inflammatory response is initiated by complete Freund's adjuvant (CFA), and an even more slowly developing inflammatory response is driven by the compound monoiodoacetate (MIA) that kills cartilage chondrocytes.<sup>8,9,67</sup> While each of these chemical models is associated with robust responses in the joint tissue<sup>62,67,68</sup> and nervous system,<sup>69–72</sup> their relevance to events underlying the initiation or maintenance of TMD pain is questionable. On the other hand, one of the most pathophysiological relevant chemical models involves the injection of the neurotrophic protein nerve growth factor (NGF).<sup>73</sup> Unfortunately, clinical trials with anti-NGF antibodies for the treatment of pain associated with osteoarthritis were suspended and then ultimately terminated because of adverse effects.<sup>74</sup> Thus, more research using NGF on the TMJ is required to understand all the types of cells NGF impacts and the molecular mechanisms affected.

**Functional jaw manipulations and mechanical stimulation.** Because abnormal forces generated by occlusal changes or prolonged, excessive jaw opening may lead to the development of TMDs in a subpopulation of patients,<sup>75</sup> preclinical models recapitulating these issues have greater construct validity than the chemical models of TMDs. Changes in mechanical loading, often referred to as mechanical perturbation, can be induced through various means such as bite raising, splinting, or repeated maximal mouth opening. Joint degeneration has been observed in most mechanically-induced models within six weeks.<sup>44,76,77</sup> What is striking about these models is that while changes within the joint and associated musculature are robust, evidence of mechanical

hypersensitivity comparable to that reported by patients with a TMD is relatively limited. This is consistent with the fact that there is a poor correlation between radiological evidence of joint damage and pain in patients with a TMD. However, it limits the utility of these models in identification of the drivers for TMD pain. Indeed, whether changes in loading plays a significant role in the development of myofascial pain in TMD remains uncertain.<sup>78</sup> On the other hand, there may be more hope for models of TMDs involving prolonged or repeated jaw opening, which has been repeatedly associated with increased mechanical sensitivity in the face and elevated inflammatory cytokine levels in the trigeminal ganglion and upper cervical spinal cord.<sup>79</sup> Interestingly, neck inflammation further heightens facial mechanical sensitivities caused by prolonged jaw opening.<sup>80</sup> This suggests that chronic tension in the neck and shoulder muscles may contribute to persistent nociception in deep craniofacial tissues, which is characteristic of patients with TMD. Nevertheless, whether findings from these models translate to humans remains to be determined.

**Surgical manipulations of the TMJ and associated structures.** Given evidence of disc and in rare cases bone damage in a subpopulation of patients with a TMD, investigators have attempted to reproduce this type of injury with surgical procedures such as disc perforation, disc displacement, discectomy, and condyle fracture to model TMJ osteoarthritis. As with non-surgical altered loading models, these surgical models are associated with robust changes in the joint.<sup>81–83</sup> Similar approaches have been used in larger species such as minipigs, which are also associated with robust changes in joint tissues.<sup>84,85</sup> Mandibular condyle fracture models have also been developed in a variety of species ranging from sheep to rats.<sup>86–90</sup> And like disc manipulations, bone manipulations are associated with robust structural changes. Unfortunately, the presence of nociceptive changes in these models has not been well characterized and as with the altered loading models there does not appear to be a direct cause and effect relationship between structural changes within TMJ and the presence of pain. These models have limited ecological validity.

A subgroup of patients with TMDs experience masticatory muscle hypersensitivity. To study masticatory myofascial pain, a model was developed by ligation of the tendon of the anterior superficial part of the rat masseter muscle (TASM).<sup>91</sup> Histopathological analysis showed inflammatory cells cluster around the injured tendon.<sup>91,92</sup> In addition, clusters of inflammatory cells mixed with tenocytes were found adjacent to the traumatized segment and infiltrated leukocytes were also seen in the muscle adjacent to the tendon. Ligation injury of the TASM in rats led to long-lasting and constant mechanical hypersensitivity (>2 months) of myogenic origin. This model might be particularly useful for studying myogenic types of TMD pain, as well as pain associated with a TMD that develops with excessive yawning or blunt force trauma.

**Genetic modification.** Mouse genetic models have been used to investigate the molecular mechanisms underlying the development and degeneration of the TMJ, as reviewed elsewhere.<sup>55</sup> Mice with spontaneous or targeted mutations in multiple genes naturally develop TMJ degeneration in a non-invasive manner, providing insights into the roles of matrix proteins, transcription factors, and growth factors. Mutations in genes encoding collagen matrix proteins, such as Col2a1 and Col11a1, led to condylar cartilage abnormalities and TMJ degeneration.<sup>93,94</sup> Transgenic mice expressing deletion of Col2a1 and double mutations of proteoglycan genes (Bgn and Fmod) exhibit progressive TMJ degeneration.<sup>95,96</sup> Similarly, a proteoglycan 4 knockout mouse model displayed abnormal cartilage, and osteophyte formation in the glenoid fossa.<sup>97</sup> Discoidin domain receptor-1 knockout mice exhibit bone surface fissures, proteoglycan degradation, chondrocyte cluster development, and other TMJ osteoarthritis degenerative changes.<sup>98</sup> These genetic mouse models are valuable for identifying the molecular factors contributing to TMJ degeneration, which appear to mirror the natural progression seen in humans. However, most studies tend to focus solely on the time course of joint degeneration and the associated biochemical changes, often neglecting evidence of altered nociception.

**Gene-environment interaction.** One of the more important discoveries from the OPPERA study was the link between the emergence of a painful TMD and abnormalities in pathways that regulate catecholamine bioavailability. Patients with a painful TMD have increased levels of circulating catecholamines<sup>99</sup> and reduced levels of catechol-O-methyltransferase (COMT),<sup>100,101</sup> a ubiquitously expressed enzyme that metabolizes catecholamines.<sup>102</sup> Functional variants in the *COMT* gene that result in low activity of the corresponding enzyme<sup>100,103,104</sup> are associated with increased onset of painful TMD and co-occurring pain conditions such as fibromyalgia.<sup>105,106</sup> In patients with TMD, a specific *COMT* polymorphism was associated with increased pain sensitivity, prolonged chronic pain, in addition to dysfunction in key brain regions related to pain processing, including alterations at the molecular level, such as local  $\mu$ -opioid receptor availability.<sup>107,108</sup> The effect of *COMT* genotype on TMD pain appears to be enhanced by environmental factors, such as stress<sup>5</sup> and injurious surgical procedures (e.g., molar extraction).<sup>109,110</sup>

Building on this clinical work, researchers established a mouse genetic and environmental approach based on the *COMT*+/- heterozygote (that produces less COMT). These mice are indistinguishable from wild-type mice until they are exposed to a three-day swim stress paradigm followed by molar extraction surgery.<sup>111</sup> *COMT*+/- mice exposed to the stress and surgical intervention exhibited orofacial mechanical allodynia and hyperalgesia of greater magnitude and longer duration. A sexual dimorphism is also observed in this model, with females exhibiting hypersensitivity to mechanical stimuli at least two weeks longer than males. Similar to patients with TMD,<sup>7,112-114</sup> *COMT*+/- mice stress + surgery intervention exhibit widespread hypersensitivity (paw, abdomen, and back) as well as changes in behavior consistent with comorbid depression and anxiety. Furthermore, *COMT*+/- stress + surgery intervention mice have increased plasma levels of the pro-inflammatory cytokines IL-6 and IL-17A, which was validated in a clinical cohort of patients with a TMD and/or other chronic primary pain conditions, including fibromyalgia, vestibulodynia, irritable bowel syndrome, and episodic migraine. Thus, this mouse model reliably recapitulates clinically- and biologically-relevant features of TMDs and may be implemented to further test underlying mechanisms and new therapeutics. In contrast to the genetic models described above, however, histological

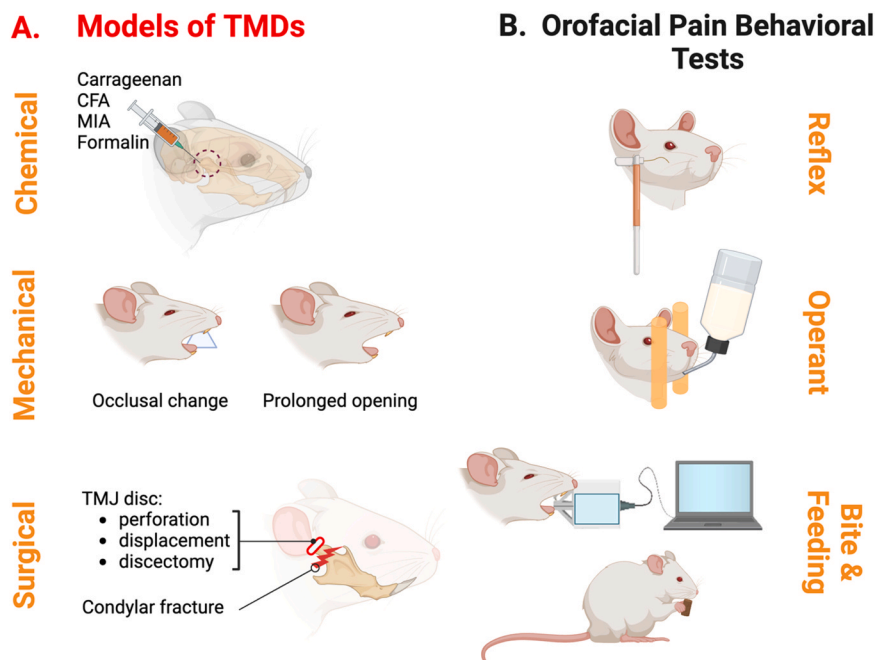
features of the muscle and TMJ have yet to be examined.

**Psychological and social models of TMDs.** With the growing appreciation of psychological and social influences on the development of pain syndromes in general and TMDs in particular, investigators are attempting to include these factors in preclinical models. One such strategy is based on evidence that stress contributes to the development as well as maintenance of TMDs. Indeed, there is evidence that orphan rats housed in single cages and exposed to chronic unpredictable mild stress exhibit increased masticatory muscle function and abnormal TMJ movement, leading to degeneration of the TMJ disc and condyles.<sup>9</sup> The isolation of the rats used in this study is just one attempt to model the impact of social interactions (or lack thereof) on the emergence of TMD symptoms. This has been replicated in multiple studies demonstrating that chronic stress exposure leads to degenerative changes in the cartilage and condyles of the TMJ.<sup>115-118</sup>

**Assessment of pain-like behaviors in preclinical models of TMDs**

*This section highlights assays used to assess pain-like behaviors in animal models of TMDs.*

Reliably evaluating behavioral responses to injury or inflammation in the masticatory system is an important aspect of successfully translating preclinical studies. Numerous behavioral assays have been developed that are optimized to enable the detection of changes in the response to stimuli in and around the joint, where evidence of increases in sensitivity is generally interpreted as evidence of TMD pain (Fig. 2). For example, sensitivity to mechanical stimuli impacting the joint or associated musculature and/or joint movement is a hallmark of TMDs.<sup>64,119-123</sup> Similarly, and even more importantly for the interpretation of changes in rodent behavior, functional behavioral changes are observed in patients with a TMD, such as limiting food intake and adjusting food quality (e.g., hard vs. soft), due to pain and dysfunction. Nevertheless, it is important to emphasize the need for caution in the interpretation of changes in rodent behavior, as it is still not possible to know what a rodent is feeling, and there are always other factors, such as changes in vigilance or hedonic drive that may influence behavior to an extent comparable to that associated with changes in nociception. With that in mind, the following are commonly used behavioral methods for



**Fig. 2.** Common TMD models (A) and assays of hypersensitivity (B) in rodents. Created in BioRender. Neubert, J. (2025) <https://BioRender.com/8aqf5tn>

assessing changes in sensitivity consistent with the presence of TMD pain in preclinical studies.

**Von Frey test for mechanical sensitivity.** Patients with a TMD rarely complain of increased spontaneous sensitivity to the skin overlaying the TMJ or associated musculature, but they will gently rub the skin overlaying these structures to alleviate movement-evoked pain. In fact, there is at least one report of a human model of TMJ pain associated with electrical stimulation of the joint detailing a decrease in the sensitivity to mechanical stimulation of the skin.<sup>124</sup> This is, of course, in marked contrast to neuropathic pain, such as that associated with small-fiber neuropathy or chemotherapy-induced peripheral neuropathy, where there is evidence of cutaneous mechanical hypersensitivity in both human patients,<sup>125</sup> and rodent models.<sup>126</sup> This hypersensitivity is easily detected and quantified in both neuropathic pain patients and rodent models of neuropathy with von Frey hairs, calibrated nylon filaments used to apply punctate stimuli to the skin. Nevertheless, researchers have consistently turned to the von Frey test to assess altered nociception in preclinical models of TMD.

When used with models of TMDs, the von Frey test is applied over the joint or muscles of mastication. Commonly used endpoints include head withdrawal,<sup>127</sup> although aggressive behavior toward the stimulus has also been scored.<sup>128</sup> Additional challenges associated with the use of von Frey hairs to assess mechanical hypersensitivity associated with TMJ injury is that in addition to aggressive behavior, where rodents attempt to bite the filament, they may also exhibit evasion behavior or swipe it with their front paws upon seeing the filament approach. To mitigate these challenges, partial restraint with hands during the von Frey test has been employed.<sup>129</sup> Alternatively, using custom chambers or devices to restrict animal's movement during the test proves to be relatively convenient.<sup>130</sup> Though it is important to note that other restraint methods may induce stress, and differences in mechanical testing procedures may potentially confound test results. A decrease in the intensity of the stimulus required to evoke a response and/or an increase in the frequency of responses is interpreted to reflect an increase sensitivity or pain. Thus, despite the clear lack of construct, if not predictive validity, the von Frey test remains in active use to assess changes in mechanical sensitivity in rodent models of TMDs. And whether because of the spread of the inflammation associated with the particular model of a TMD condition or a species difference in the neural circuitry underlying the emergence of central sensitization and referred pain, the presence of mechanical sensitivity as detected with von Frey hairs has been reported in various models of TMDs, such as capsaicin, CFA, MIA, TASM, and sustained and repeated maximal mouth opening.<sup>69,70,91,131–133</sup> That said, investigators have attempted to modify the von Frey hair test to make it more appropriate for the stimulation of deeper structures, in a manner comparable to the pressure algometers used in humans. This has been done by desensitizing rodents to cutaneous stimulation as well as the use of a blunt tip, to facilitate the application of forces to deeper structures.<sup>134</sup>

**Grimace scale.** Facial expressions in response to noxious stimulation are stereotypical across species, including humans. Consequently, facial expressions have been used to infer the presence of, if not quantify the magnitude of acute pain in humans, where it has been used most extensively in neonates.<sup>135,136</sup> Mogil and colleagues were the first to propose a formal scale for the quantification of what they referred to as facial expressions of pain in the mouse<sup>137</sup> that involved assessment of five facial features (orbital tightening, nose bulge, cheek bulge, ear position, and whisker position) scored on a three-point scale (not present, moderate, and severe). While originally developed to detect the presence of headache, the scale has been subsequently used for inferring the presence of spontaneous or ongoing pain in a growing number of models,<sup>137–139</sup> including models of TMDs<sup>131,140–143</sup> where it has been used to detect changes at least soon after the generation of the model. Like in humans, however, where facial expression becomes a poor reflection of pain, the utility of grimace to infer the presence of pain in chronic models is more limited. Furthermore, and potentially more

problematic for the use of the grimace scale in models of TMD, is that it is optimized for the detection of spontaneous or ongoing acute pain, while TMD pain is most prominently elicited during functional use of the TMJ. There are, of course, several other stimuli that drive changes in the five facial features monitored, for example a sleeping mouse may be scored as severe on all five features. The initial application of the scale was also highly subjective on the part of the scorers and scoring was extremely time consuming, as single frames from video recordings were needed to be captured, cropped, and scored. And while these last two issues have been recently addressed with an automated artificial intelligence-based approach to scoring videos called PainFace (<http://painface.net>),<sup>144</sup> the predictive validity of this endpoint in the context of TMDs has yet to be demonstrated.

**Food intake and TMD functional assessment.** The clinical pain research community has gradually shifted away from relying solely on numeric pain intensity ratings, recognizing that more natural endpoints involving activities of daily living may better reflect the efficacy of therapeutic interventions and hold greater relevance for patients. Similarly, preclinical pain researchers have begun incorporating comparable endpoints into preclinical models. Furthermore, given that a dominant clinical characteristic of TMD is functional pain, such as during chewing, meal pattern analysis has proven to be a sensitive measure of loss of function, if not discomfort, in preclinical models of TMDs. Additional advantages of meal pattern analysis are that it is an objective measure, relatively easy to implement, and particularly useful with chronic disease models. An increase in meal duration is observed in the CFA model of TMDs which is more robust in females than males.<sup>145–147</sup> Importantly, the increase in meal duration is observed in conjunction with decreased cartilage thickness, tissue swelling, synovitis, and inflammation in retrodiscal tissue.<sup>145–147</sup> Nevertheless, while changes in meal pattern have face and construct validity, there are several factors independent of pain, such as TMJ dysfunction or a more generalized illness behavior, that may influence meal patterns and food intake in humans with a TMD. Additional caveats associated with this endpoint include the need for specialized equipment, (such as a nose poke system for food pellet delivery, so that it is possible to monitor meal duration and volume), the need to single-house animals on a wire mesh floor (both of which may be stressful, where single housing is even more stressful in females), and the need for a relatively stable model so that meal pattern can be monitored over several days.

**Operant assays.** Operant assays, as the name implies, require the animal to act on its environment in some way. These actions or behaviors may be as simple as moving from one side of a testing box to another. Thus, these assays often involve some degree of learning and when used in the context of pain models, often involve a choice, such as between a nociceptive stimulus and access to a reward or between a nociceptive stimulus and some other unpleasant stimulus such as a bright light. Consequently, these assays are also described as reward/conflict assays. These assays also differ from other stimulus-response assays in that they allow the animal to regulate the degree of nociceptive stimulation. These paradigms are especially useful for drawing comparisons with human studies as they allow animals to select their own response strategy. Importantly, operant behaviors are characterized by their intentionality, motivation, and learned nature, and they typically involve complex “higher” processing which has obvious generalizability to what is known about pain processing in humans.

Several operant assays have been developed to assess oromotor function and/or changes in sensitivity of perioral structures including the Dolognawmeter, and the orofacial pain assay or OPAD. These assays vary in their ease of use and relevance to TMDs, but all have been used with TMD models to support evidence of hypersensitivity. Of note, while operant assays may provide greater insight into the complex psychobiological processes underlying pain and avoidance strategies, the interpretation of the changes observed should be made even more cautiously as the number of potentially confounding variables increases the farther the endpoint in question is from a direct response to noxious stimulation.

**Bite force measurement for masticatory pain.** Under most normal circumstances, mastication as a component of instinctive behavior integral to food consumption is not perceived as painful. However, for some patients with a TMD, mastication may become very painful. This can be understood as a specific case of masticatory allodynia, which eventually leads to reduced bite force. Clinical research indicates that patients with a TMD have significantly reduced bite force compared to healthy controls, with this reduction inversely proportional to pain severity.<sup>148–150</sup> Based on this, methods to measure bite force in mice<sup>143,151,152</sup> and rats<sup>153</sup> as a clinically relevant metric of TMD masticatory pain have been developed. A significant reduction of bite force was observed for up to nine days in the CFA model and 21 days in the TASM model in mice.<sup>152, 154,155</sup> Using this approach it was reported that TRPV1, TRPA1, TMEM100 (a regulator of TRPV1-TRPA1 interaction), and TRPV4 are involved in TMD masticatory pain.<sup>24,152,154–156</sup> The Dolognawmeter,<sup>157</sup> developed by Schmidt and colleagues, is another assay that directly assesses changes in oromotor function. The design is simple, elegant, and, like the bite-force assays, provides an objective measure of sensitivity that can be repeatedly assessed over time. The assay consists of a tube with two blocking dowels between the mouse and a reward. A timer is triggered after the mouse chews through the first dowel and is stopped when the mouse chews through the second. Thus, the latency to chew through the second dowel is the dependent variable used to quantify changes in sensitivity.<sup>157</sup> However, because of the use of the tongue to move the chewed material out of the mouth, the assay appears to be most robust for the detection of changes in tongue sensitivity. Nevertheless, it does appear to enable detection of sensitivity in the TMJ and muscles of mastication.

**Orofacial Pain Assay (OPA) and the OPAD.** Two additional devices enable detection of changes in perioral sensitivity. Both devices are commercially available and involve the same basic paradigm, where access to a reward such as condensed milk or a sucrose solution requires the mouse or rat to tolerate mechanical or thermal stimulation of the whisker pads or muscles of mastication. This is achieved by lining a portal the rodent must use to access the reward with the appropriate stimulus. The assays are relatively easy to use and enable quantification of several different variables including nose pokes through the portal and licking time. A ten-minute test session is sufficient to detect dramatic changes in sensitivity, such as that associated with constriction of the infraorbital nerve.<sup>158</sup> Because the stimulation devices may be removed, the assay enables the assessment of hedonic drive (reward seeking behavior) in the absence of nociceptive stimulation, which is a critical control for all of the reward-based assays given evidence that chronic pain is associated with depression, and depression is associated with a decrease in hedonic drive. These assays have now been used to assess therapeutic efficacy in a number of orofacial pain models.<sup>159–169</sup> While the devices are designed to most effectively stimulate superficial structures, they do appear to facilitate the assessment of muscle and joint nociception as well and have been successfully used in a model of TMDs.<sup>170</sup>

## Tissue engineering and computer modeling approaches to studying and treating TMDs

*This section focuses on tissue engineering, computational, and functional analyses of TMDs.*

**Tissue engineering approaches to treating joint damage TMDs.** With evidence of disc damage in a subpopulation of patients, several investigators have leveraged recent advances in tissue engineering to develop novel therapeutic approaches for treating the damaged TMJ. The goal is to replace damaged tissue with healthy engineered tissue. In one such approach allogeneic costal chondrocytes were used to create transplantable, cell-based, scaffold-free TMJ implants.<sup>84</sup> Larger animals such as the minipig, pig, goat and sheep have been used for these approaches because of similarities to humans in TMJ tissue mechanical properties, diet and chewing patterns, TMJ physiology, and disease

progression.<sup>44</sup> In the minipig model, constructs were implanted into partial thickness focal defects of the TMJ disc using an intralaminar fenestration technique to secure TMJ neodisc constructs between two TMJ disc laminae. Over an eight-week period, these implants showed robust integration and superior mechanical strength compared to untreated defects. Importantly, treated animals exhibited fewer instances of pathological or abnormal condylar remodeling, highlighting the therapeutic potential and providing proof-of-concept for a cell-based, tissue-engineered disc replacement.

An alternative acellular approach focused on extracellular matrix (ECM) derived from small intestine submucosa (SIS) to remodel into a disc-like tissue was studied in pigs, canines, and goat. In initial farm pig study,<sup>171</sup> rapid remodeling of SIS implants was observed, with the tissue resembling native TMJ disc by one month and integrating with the original peripheral disc attachments. In a subsequent translational study, studies have tested SIS implants in a canine model.<sup>172</sup> This good laboratory practices (GLP)-compliant study confirmed the safety of the ECM devices, produced using good manufacturing practices (GMP)-like methods, with no adverse effects observed, further supporting their potential for clinical translation. While promising results were obtained with ECM implants in minipig, goat and canine models, other studies using synthetic interpositional implants made of synthetic polymers in sheep faced significant challenges. Despite rigorous efforts, these studies resulted in device failure and degenerative changes in the joint space,<sup>173</sup> highlighting the complexities and species-specific responses in TMJ implant development. These tissue engineering studies underscore the potential and challenges of developing TMJ disc implants. These findings are crucial for advancing TMJ disc replacement therapies and underscore the importance of continued research and refinement in this field.

Tissue engineering-based strategies are also in development as solutions for *TMJ condylar replacement*. One recent approach involved fabricating load-bearing scaffolds for condylar replacement, demonstrating that cellularized scaffolds can support osteochondral tissue development. In a pilot study,<sup>174</sup> a load-bearing polycaprolactone (PCL) scaffold for condylar resurfacing was tested in minipigs. The scaffold featured a long-slotted shaft for fixation on the ramus and was seeded with autologous mesenchymal stem cells (MSCs) derived from the iliac crest. Results showed significant osteogenesis in the condyle head, with the formation of cartilage pockets on the articular surface. This study provided promising proof-of-concept for using cellularized PCL scaffolds in TMJ condylar replacement.

An alternative approach was based on ex vivo tissue engineering of TMJ condylar bone.<sup>175</sup> Addressing the clinical need for condyle regeneration, xenografts were milled from bovine decellularized bone to achieve human anatomical shape, then these scaffolds were seeded with progenitor cells. The constructs were cultured for five weeks in a customized perfusion bioreactor, essential for proper tissue formation by ensuring nutrient and waste transport and providing mechanical stimulation via hydrodynamic shear. Whether the tissues grown in vitro can be successfully transplanted remains to be determined. Furthermore, while all four tissue engineering based approaches described are yielding promising results, none of these have included nociceptive endpoints in the analyses. With evidence of a dissociation between structural changes in the joint and the magnitude of associated pain, it remains to be seen whether either of these disease modifying approaches will influence TMD pain.

**Computational Models of the TMJ.** Computational modeling has become an important tool in preclinical TMD research. The potential to easily manipulate the structural or functional set-up and facilitate the investigation of “what-if” scenarios, that are either technically or ethically not feasible in animals or humans, makes computational models a unique instrument for TMD research. Previous computational modeling work of the TMJ can be roughly categorized into two groups: (1) multibody and (2) deformable models.

**Multibody Models.** From a historical perspective, in silico modeling of

the TMJ has focused on muscle driven, rigid body models. These models have facilitated understanding of reaction forces in the TMJ during tasks like chewing or mouth opening.<sup>176,177</sup> More recent, multibody models of the jaw have used bi- or unilateral point constraints,<sup>176,178</sup> elastic foundation contact models<sup>179</sup> or sphere-plane contact models.<sup>180</sup> While these multibody models have been useful for estimating forces that may be generated within the joint, the clear drawbacks are the use of non-deformable structures and lack of a TMJ disc, which limits the utility of the approach for detailed investigation of TMJ biomechanics, and in turn mechanobiology.

**Deformable models:** The use of finite element (FE) modeling to represent deformable TMJ tissues has expanded considerably in recent years. In material modeling, research has characterized the biphasic behavior of TMJ disc tissue<sup>181–184</sup> along with solute diffusion properties,<sup>185</sup> electrical conductivity,<sup>186</sup> charge density,<sup>187</sup> and nutrient environments.<sup>188</sup> This added complexity generally increases computational costs, limiting most applications of these models to static tasks like clenching.<sup>189,190</sup> However, some dynamic FE models exist,<sup>191,192</sup> and with rapid increases in computational power, the use of dynamic models in TMJ research will likely expand. One way to reduce computational requirements is to use a combined multibody-FE approach. This is typically done by using the FE method for the TMJ while modeling bones as rigid bodies and muscles as linear actuators.<sup>193,194</sup> Recently, this approach was used to study the impact of morphological changes on TMJ loading during dynamic lateral bruxing<sup>195,196</sup> and the effect of food bolus variables on TMJ biomechanics during chewing.<sup>197,198</sup> This technique has advantages over co-simulation, which uses a multibody model to compute time-step input forces for quasi-static FE simulations. By requiring only one simulation toolkit, it reduces setup complexity and time while ensuring that forces and mechanical stress on FE components are computed within the same simulation. This approach avoids errors that may arise from using forces and motions derived from simpler multibody models, which may not align with the more complex contact-based FE joint setup.

Computational models of the masticatory region have greatly advanced understanding of how morphological variables impact joint loading and potential injury development. They have also clarified connections between functional and parafunctional tasks and TMJ loading. Although computational animal models of the masticatory region exist,<sup>199–202</sup> they generally do not focus on the TMJ. While human models are advantageous for clinical translation, more complex modeling strategies (such as advanced material properties or the inclusion of mechanobiology and cartilage breakdown) require detailed input and validation data (e.g., needle EMG, implanted EEG, implanted force sensors). Since such studies cannot be conducted ethically in humans, developing high-fidelity computational animal models of TMJ biomechanics is essential to understanding mechanobiological processes related to pain development and disease progression.

**Neuroimaging and needed advancements in preclinical TMDs research.** In contrast to human neuroimaging research on TMDs, studies using pre-clinical neuroimaging in models of TMDs are currently much more limited. Research using fMRI in a rodent model of irritable bowel syndrome (IBS) and myofascial-like pain (induced by stress after masseter muscle injection) shows that brain regions, including the insular cortex and reward/limbic systems, exhibit higher responses to evoked visceral pain.<sup>203</sup> However, compared to a stress-induced IBS-like pain model that produces visceral hypersensitivity for days to weeks (longer in males than females),<sup>204</sup> it is notable that the combination of IBS-like and myofascial-like pain leads to much longer durations of visceral hypersensitivity (i.e., on the order of months).<sup>203,205</sup> Additionally, a literature search for preclinical neuroimaging of TMDs-like models identified only one published study involving a comorbid TMDs-pain model.<sup>203</sup> While preclinical neuroimaging research holds promise for establishing causative links between changes in the brain observed in human studies of TMDs and the signs and symptoms of TMDs, the choice of a model and the connections between the model chosen and the subpopulation of

patients with a TMD being modeled will be essential to resolve.

## Integrated physiology approach in TMJ research

*This section presents a reflective perspective on integrating physiology into TMJ translational research.*

In most degenerative joint diseases, there is often a significant misalignment between the severity of structural changes and the severity of symptoms. As noted above, this is also true for TMDs involving the joint, where joint pathology does not necessarily correlate with patient-reported symptoms or disability. Compounding the challenge is the fact that TMDs encompass a range of pathologic conditions, including TMJ degeneration, changes in masticatory muscle function, and inflammatory conditions, which occur in the majority of subjects alongside COPCs. Since patients with a TMD may be seeking treatment for chronic joint pain and disability, it is crucial to understand the pathological features that lead to chronic TMJ pain and how these features differ among patients. This knowledge is essential for identifying the most appropriate treatments for individual patients. To advance TMDs research and develop the next generation of treatments, it is necessary to stratify patients into TMD biopsychosocial endotypes. To achieve this, deep pain phenotyping and use of calibrated assessment tools, such as the DC/TMD (Diagnostic Criteria for Temporomandibular Disorders), can provide detailed information on the patient's sensory function, masticatory system function, pain experience, sleep quality, coping skills, emotional support systems, and many other factors. This stratification will help precisely identify modifiable features within these cohorts, leading to improved masticatory system function and reduced pain for patients. Once stratified into defined subgroups based on the characteristics of TMD pain, reverse-translation should be possible, where targets can be identified and novel interventions tested. The goal is to identify interventions that are most likely to benefit a specific patient based on their individual pain experience, biology, and physiology, psychology, social structure etc. whether those treatments are pharmaceutical, rehabilitative, behavioral, dietary, or surgical (see Table 1).

Unfortunately, there are gaps in this approach that need to be addressed. First, while we have begun to identify pain-associated subgroups in patients with a TMD, much larger datasets are necessary, along with measurements of physiological systems that were not fully characterized in OPPERA. The well-designed structure of OPPERA significantly advanced our understanding of TMDs and sets the stage for productive next directions. Thus, data integration across multiple studies and research sites is essential to assemble the puzzle pieces in a meaningful way. Efforts toward FAIR (findable, accessible, interoperable, and reusable) data practices, along with the use of common data elements (CDEs), have helped identify integration points between studies and across various sites. However, achieving perfect alignment of data and methods across studies presents a significant, if not insurmountable, challenge. Therefore, alongside FAIR data practices, there is a need for transparency and documentation of data collection practices. The publication of protocols and version control through online publishing services can help clarify how and why data discrepancies arise between studies. Moreover, the preservation of samples through biorepositories linked to deep pain phenotyping data allows for the dataset to be 'widened' over time without the need for repeated pain phenotyping. By creating thousands to millions (or more) data points within the same subjects, emerging data science techniques can be employed to stratify patients into unique subgroups.

## Discussion

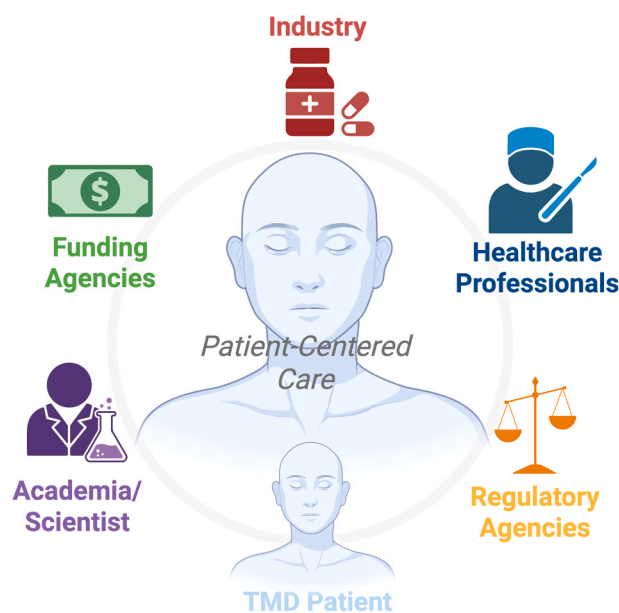
*A patient-centered TMD collaborative (Fig. 3)*

At the recent 2024 International Association for Dental Research (IADR) session in New Orleans, a pre-meeting was convened by TMD

researchers interested in forming a TMD Collaborative to advance research in this area. These researchers were represented by a breadth of teams from diverse institutions including: Clemson, Duke, Harvard University, HealthPartners Institute, Missouri State University, Stanford, Texas A&M University, University of British Columbia, University of Buffalo, UC Irvine, UCLA, UCSF, UNC-CH, University of Florida, University of Michigan, University of Pittsburgh, University of Southern California, University of Texas at Health Science Center at Houston and Massachusetts General Hospital. There was general agreement among all teams with the National Academies of Science, Engineering, and Medicine (NASEM) Report,<sup>1</sup> that highlighted the fact that despite considerable investments of both public and private funds to mitigate the deleterious impacts of TMDs on patients, families, and the broader community, funding to date has been insufficient to achieve this goal. Consequently, effective interventions remain a goal for clinicians and patients alike. Numerous reasons contribute to the limited efficacy of even symptom management strategies, including the lack of phenotypic data on patients with a TMD, heterogeneity in the manifestations of these complex disorders, and the impact of COPCs. Recently, however, the standard approach employed by the biomedical research community to identify and develop novel therapeutic interventions for pain syndromes, particularly TMDs, has come under scrutiny. There was also a consensus that there are issues with the models, endpoints, and possibly the species employed. Despite the limited understanding of the mechanistic drivers of pain, functional limitations, and impairment in patients with a TMD, there is a growing appreciation for the fact that injecting a noxious inflammatory substance into the joint space to induce experimental pain lacks face or construct validity. In short, such experimental model approaches have poor ecological validity and do not appear to reflect mechanisms driving patients with a TMD to seek medical attention. Similarly, while changes in sensitivity to cutaneous thermal and mechanical stimuli are robust in preclinical models, they are largely absent in patients with a TMD. In other words, the complexities of persistent musculoskeletal pain transcend the oversimplification of these experimental models. Consequently, while all TMD Collaborative for Improving Patient-Centered Translational (IMPACT) research teams acknowledged the importance of preclinical models in finding viable solutions to this debilitating problem, they all agreed that new approaches are needed for these models to yield pivotal clinical insights.

A two-prong approach was proposed to enable the more effective management of TMDs. The first prong involved the development of a coordinated patient-centered approach (Fig. 3). Coordination in this case was within and between all relevant parties including patients, healthcare providers, payers (insurance companies), funders (NIH), regulatory agencies (FDA), and researchers. Effective communication is essential in addressing this unmet need, where inter- and multidisciplinary teams will be critical to overcoming the current issues, paving the way for establishing evidence-based treatment modalities of TMDs. The second prong leverages the foundation laid by the OPPERA studies, which was further refined through the strategy employed in the back pain consortium (BACPAC).<sup>206–208</sup> Data derived from in-depth and comprehensive phenotyping of individuals with TMDs should not only serve as a foundational guide for the development of preclinical models (Fig. 4) but can be used to design and facilitate clinical trials, develop personalized treatment strategies, and guide diagnosis and prognosis of patients presenting with a TMD. Thus, in addition to the psychophysical, psychometric, and genetic data collected in OPPERA, it is now possible to not only collect but analyze a far richer data set from each patient, ranging from a full genome analysis to patient reported assessments over time in their natural environment. Furthermore, high resolution imaging can help identify changes in pain-inhibitory circuits and signaling molecule systems in patients with a TMD.

Detailed functional analyses are now possible, as are potentially disease modifying factors like the oral and gastrointestinal microbiome, sleep quality, and social interactions. Powerful analytic tools including machine learning and artificial intelligence can facilitate patient



**Fig. 3.** Visual summary of the key elements in a holistic approach to achieving better patient-centered care of patients with a TMD.

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stratification. This can also be used for refined computer modeling and joint structure and function. The factors used to stratify patients can be used to guide hypothesis-testing experiments designed to distinguish biomarkers from causal mechanisms as well as the development of more refined and appropriate preclinical models with which to establish causal relationships. If patients are followed over time, treatment and outcome data can be included in the models not only facilitating diagnosis but prognosis. The ideal of course is the development of individualized treatment approaches.

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#### Declaration of Generative AI and AI-assisted technologies in the writing process

During the preparation of this work, the authors used ChatGPT (GPT-4, OpenAI) to enhance the clarity and consistency of the content. After using this tool, the authors reviewed and edited the content as needed and take full responsibility for the content of the publication.

#### Author contributions

J.K.N. and M.S.G. conceptualized the article. All authors contributed to its academic content and discussions. A.A., P.D., and N.P.M. revised and edited the manuscript prior to submission. All authors reviewed and approved the final manuscript.

#### Declaration of competing interest

The authors declare that they have no known competing financial

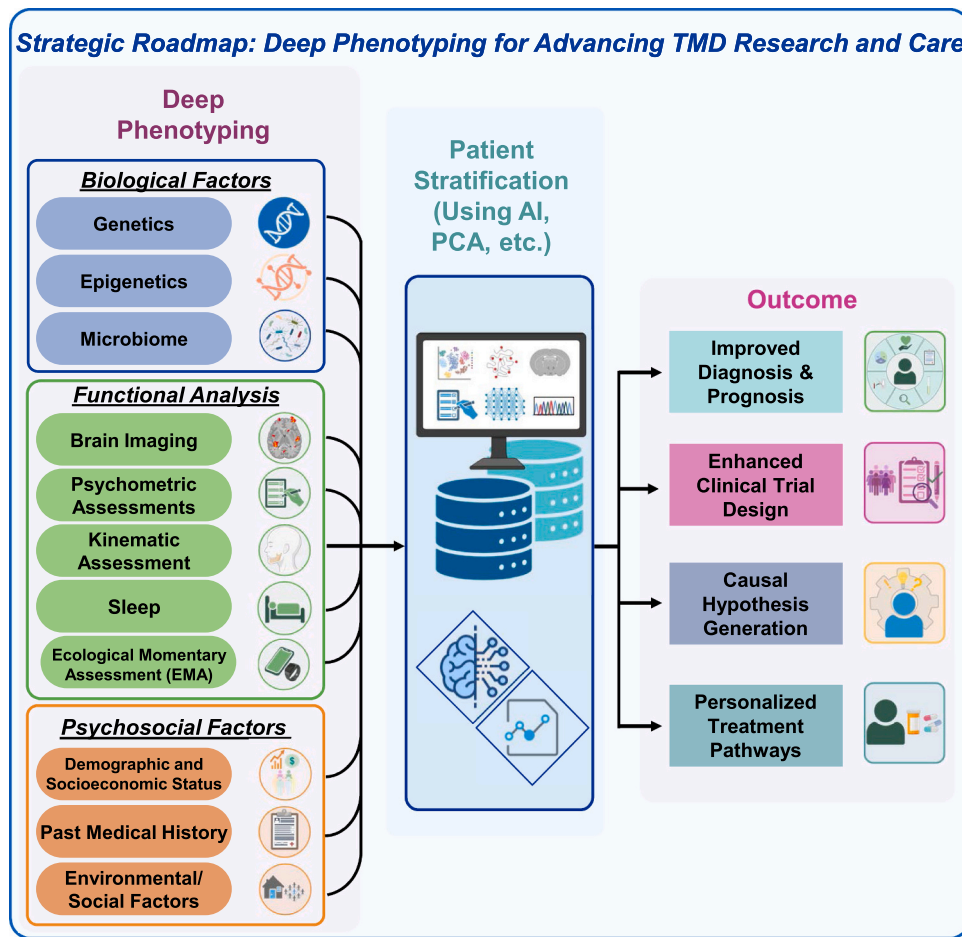


Fig. 4. Strategic roadmap - deep phenotyping for advancing TMD research and care. Created in BioRender. Reddy, A. (2025) <https://BioRender.com/9we2x1k>

interests or personal relationships that could have appeared to influence the work reported in this paper.

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