Received: 30 March 2021 | Revised: 18 May 2021 | Accepted: 9 July 2021

DOI: 10.1111/odi.13959

ORAL DISEASES Leading in Oral, Maxillofacial, Head & Neck Medicine WILEY

SHORT COMMUNICATION

Functional hypothesis of the juxtaoral organ: Role of collagen types I and III

Antonio José Mérida-García[1] | Jorge Murillo-González[2,3] | Elena Martínez-Sanz[2,3] | Javier Catón[2,3] | Luis A. Arráez-Aybar[2,3] | José Martín-Cruces[4] | Teresa Cobo[5,6] | José Antonio Vega[4,7] | José Ramón Mérida-Velasco[2,3]

[1]Servicio Oncología Médica, Complejo Asistencial, Zamora, Spain

[2]Facultad de Medicina, Departamento de Anatomía y Embriología, Universidad Complutense de Madrid, Madrid, Spain

[3]Laboratorio de Desarrollo y Crecimiento Craneofacial, Facultad de Odontología, Grupo UCM de Investigación, Investigación en Desarrollo del Paladar y Fisura Palatina. Desarrollo Craneofacial', Universidad Complutense de Madrid, Madrid, Spain

[4]Departamento de Morfología y Biología Celular, Grupo SINPOS, Universidad de Oviedo, Oviedo, Spain

[5]Departamento de Cirugía y Especialidades Médico-Quirúrgicas, Universidad de Oviedo, Oviedo, Spain

[6]Instituto Asturiano de Odontología, Oviedo, Oviedo, Spain

[7]Facultad de Ciencias de la Salud, Universidad Autónoma de Chile, Santiago de Chile, Chile

Correspondence
Jorge Murillo-González, Facultad de Medicina, Departamento de Anatomía y Embriología, Universidad Complutense de Madrid, 28040 Madrid, Spain.
Email: jmurillo@med.ucm.es

Funding information
This work was partially supported by grant number PR87/19-22587 from Santander Bank/Complutense University of Madrid.

Keywords: collagens type I/III, function, histology, immunohistochemistry, juxtaoral organ

The juxtaoral organ (JOO) was first described in human embryos by (Chievitz, 1885). In human adults, it has been described in the bucco-temporal space—located within the soft tissue between the medial surface of the mandible and the buccinator muscle—innervated by the buccal nerve (Zenker, 1953). Histologically, the JOO consists of parenchyma formed by nests of non-keratinizing squamous epithelial cells surrounded by two layers or strata of connective tissue that form the stroma—the stratum fibrosum externum (SE) and the stratum fibrosum internum (SI). In addition, the JOO is rich in small nerve branches that form the stratum nervosum (Zenker & Salzer, 1961). Some reviews have reported epithelial islands residing in peripheral nerves similar to JOO, cases of hyperplasia, hamartomas or neuroectodermal tumours; even in biopsies, the JOO can be misinterpreted as carcinomas (Pantanowitz & Balogh, 2003; D'Andrea et al., 2015; Suárez-Quintanilla et al., 2020).

The function of the JOO is not clearly defined. Despite numerous immunohistochemical studies on the JOO, there are no studies on the expression of collagen types I and III in its stroma (Table 1). Such information is crucial for its understanding since collagens play structural roles and contribute to mechanical properties, organization and shape of tissues (Ricard-Blum, 2011). Thus, the objective of this work was to carry out a histological study of JOO after dissection of the infratemporal region in adult human cadavers, analysing the immunoexpression of collagen types I and III and its disposition (by using the picrosirius red staining technique).

For this study, sixteen formalin-fixed cadaver-head specimens were collected from the former Department of Anatomy and Embryology II and the Body Donation Centre of the Complutense University of Madrid. In all cases, a bilateral dissection of the infratemporal fossa was performed. In six cases, the JOO dissection was attempted, but it could not be identified macroscopically. In 10 cases, we explanted the region where the JOO was suspected to be. Therefore, the buccal nerve and surrounding tissue were resected, from its access to the pterygomandibular region until the buccinator muscle was reach (Figure 1). Histological examination allowed the JOO to be identified. The preparations were processed

TABLE 1 Immunohistochemical characterization of the juxtaoral organ

References	Species	Positive immunostaining	Negative immunostaining	Additional information
Soucy et al. (1990),	Human	NSE & GFAP	NS	Case report of a biopsy in a 5-year-old girl
Mandl et al. (1993)	Human	CK KL1 & CK 19	Desmin, chromogranin, NSE & S–100 protein	Samples of a juxtaoral organ from biopsies. Transmission electron microscopy was also performed. Article in German
Vadmal et al. (1998)	Human	Vimentin (+++), CK CAM5.2, CK AE1/AE3 (+) & epithelial membrane antigen (+)	S–100 protein, smooth muscle actin, chromogranin, synaptophysin, NSE & GFAP	Case report of a intraoral tumour in a 12-year-old girl
Ide et al. (2004)	Human	S–100 protein, NSE, neurofilament protein 2F11, epithelial membrane antigen E29 & CKs AE1/AE3, 34bE12 & MNF116	chromogranin & synaptophysin	Case report of a biopsy in a 35-year-old man
Bahcelioglu et al. (2005)	Dog	EGF-r, TGF-α & NGF-β	NS	Authors suggested a neurosecretory function for the JOO
Kusafuka and Kameya (2007)	Human	Pan-CK AE–1/3, CK 19, high molecular-weight keratin & Melan-A	CK7, CK20, vimentin, S–100 protein, human melanosome, CEA, E-cadherin, CK14, chromogranin-A & synaptophysin	Case report of a biopsy in a 44-year-old Japanese woman
Velasco et al. (2012)	Rat embryos	HNK–1 (in E14 & E15)	HNK–1 (from E16 to E19)	Authors studied from rat development, from E14 to E19
Kobayashi et al. (2015)	Mice	CK14, NF, Ki67 (in 2 week-old mice), p63 & Glut1 (both 2 & 8 week-old mice)	CK18, Ki67 (in 8 week-old mice) & Glut1 (in 1 week-old mice)	Authors studied several developmental stages in mice
Suárez-Quintanilla et al. (2020)	Human foetus	S100 protein, NSE, CD34, PIEZO2	ASIC2 & TRPV4	Foetal tissue. Authors suggested a mechanosensory role

Note: (+) weak staining; (+++), strong staining; ASIC, acid-sensing ion channels; CKs, Cytokeratins; E, embryonic day; EGF, epidermal growth factor; GFAP, glial fibrillary acidic protein; Glut1, anti-glucose transporter1; HNK-1, human natural killer-1; NF, neurofilament; NGF, nerve growth factor; NS, Not Specified; NSE, Neuron-specific enolase; PIEZO2, Piezo Type Mechanosensitive Ion Channel Component 2; TGF, transforming growth factor; TRPV, transient receptor potential vanilloid.

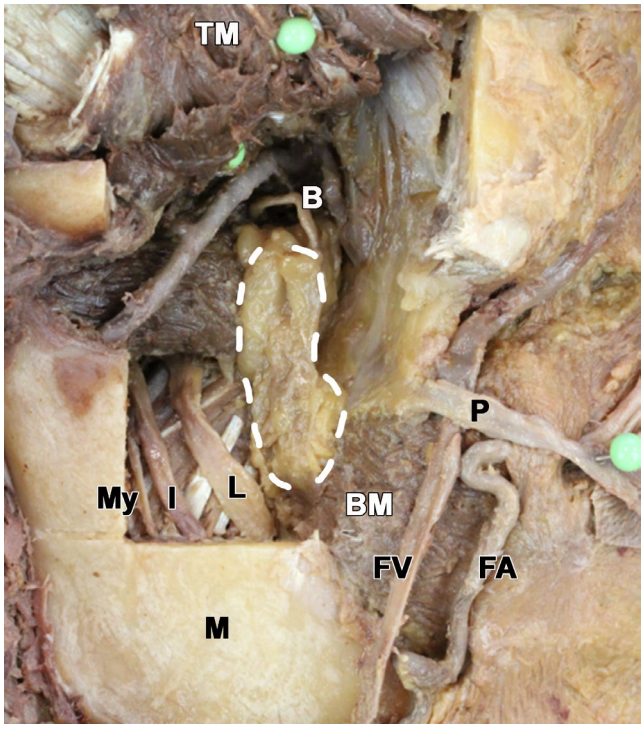

FIGURE 1 Lateral view of the dissection (right side). Aspect of the pterygomandibular region: the zygomatic arch, the coronoid process of the mandible and the temporalis tendon were cut; the dashed white line indicates the area where the tissue surrounded the buccal nerve (B) was resected. TM, temporalis muscle; M, mandible; FV, facial vein; FA, facial artery; P, parotid duct; BM, buccinator muscle; L, lingual nerve; I, inferior alveolar nerve; My, nerve branch to supply the mylohyoid muscle and the anterior digastric belly

and routinely stained, including picrosirius red. Furthermore, immunohistochemistry on paraffin sections was performed following the protocol for the ready-to-use Vectastain (PK-7200). The following primary antibodies were used: mouse anti-collagen type I (sc-59772, 1:200) and rabbit polyclonal antibody anti-collagen type III (bs-0549R, 1:200).

According to our findings, the JOO is a fusiform structure of variable length (6–11 mm) and thick (1–2 mm), surrounded by loose connective tissue (Figure 2a). The parenchyma of the JOO is formed by interconnected nests of epithelial cells, of different size and shape, with a multilobular appearance (Figure 2b-d). In some nests, the internal cells show a whorl-like or concentric arrangement and a larger cytoplasm, and other nests show small vesicles with amorphous material (Figure 2e). The stroma was formed by dense connective tissue that forms the capsule (SE), organized in concentric layers (Figure 2b,e,f), in which type I collagen predominated (Figure 2g). Inside the stroma, the SI was formed by loose connective tissue, with a predominance of type III collagen, arranged radially between the epithelial nests (Figure 2f,h).

A range of immunohistochemical findings have been reported by immunohistochemical studies performed in different species and types of tissues, focused on the parenchyma or the nerves fibres (Table 1). However, there are no previous studies of the expression of collagen I and III in the stroma that emphasize its role as a mechanoreceptor.

The role of the JOO stroma is important for its organization and morphological function. Previous studies suggested that the difference in the proportion of type I/III collagens, and the arrangement and size of the fibres are important characteristics for the mechanical resistance and elasticity of the tissues, since the elastic and dynamic properties are significantly different in both types of collagens (Asgari et al., 2017). We suggest that the SE formed predominantly by concentrically arranged collagen I, giving the JOO solidity and resistance to stretching and compression. The dense connective tissue that surrounds the small neurovascular bundles that reach the JOO serves to fix it to the neighbouring tissue. In addition, the SI principally formed by radially arranged collagen III provides the elastic properties to JOO. However, immunostaining of collagen type III is also slightly noticed in the SE. These results are consistent with the general distribution and function of collagen types (Gelse et al., 2003). Indeed, collagen type III is associated with collagen type I in many tissues (Kuivaniemi & Tromp, 2019).

The importance of the JOO stroma during development has been reported previously. In rats, there is evidence that it is derived from the neural crest (Velasco et al., 2012). Furthermore, it has also been suggested that the melanin pigmentation identified within the JOO stroma in human adults is produced by melanocytes derived from the neural crest (Ide et al., 2003). Recently, it has also been reported that the stroma presents perineural characteristics (Suárez-Quintanilla et al., 2020).

The JOO is located in the pterygomandibular region in relation to the buccal nerve. Its topographical relationships with the masticatory muscles, the buccinator muscle and the pterygomandibular raphe, as well as the organization of its stroma, suggest that JOO acts as a mechanoreceptor stimulated by actions such as chewing or swallowing.

The knowledge of the topography and histology of the JOO can help to avoid misinterpretations related to the invasive front of carcinomas. Indeed, some microscopic features have been reported to be helpful in differentiating between the JOO and invasive perineural carcinoma (Müller, 2016; Pantanowitz & Balogh, 2003). In addition, this report also suggests that the immunoexpression of collagen types I and III in the stroma of the JOO seems to be consistent with its role as a mechanoreceptor.

ACKNOWLEDGEMENTS

This work was partially supported by grant number PR87/19-22587 from Santander Bank/Complutense University of Madrid, to which we thank. However, the financial support has not influenced the results, and none of the authors discloses any conflict of interest. The authors also wish to sincerely thank those who donated their bodies to science so that anatomical research could be performed. Results from such research can potentially improve patient care and increase our overall knowledge. Therefore, these donors and their families deserve our highest gratitude.

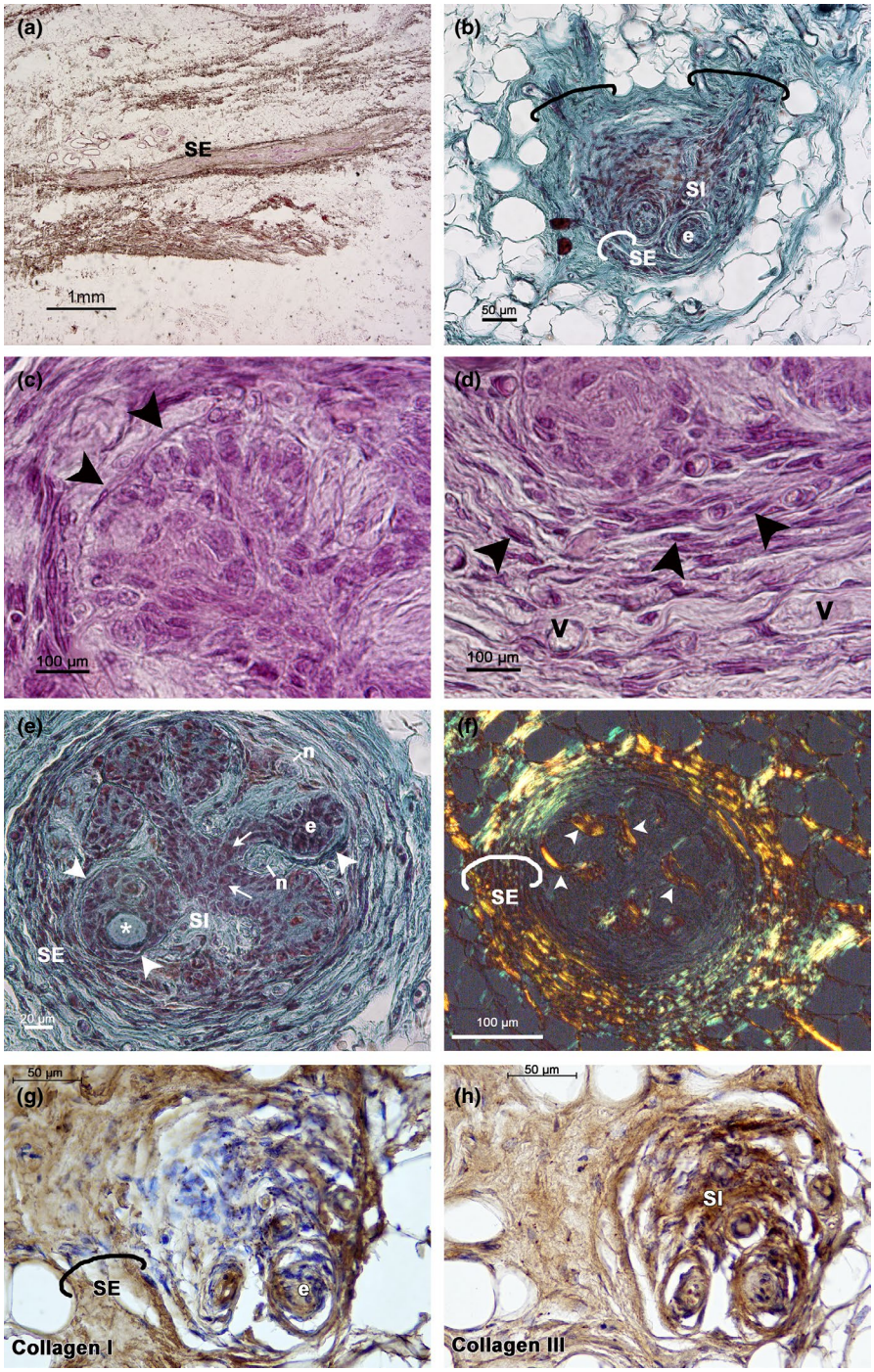

FIGURE 2 Juxtaoral organ (JOO) histology. (a) Longitudinal section of the JOO; haematoxylin–eosin staining. SE, stratum fibrosum externum. Scale bar, 1 mm. (b-h) Frontal histological sections of the JOO. (b) VOF-III staining showing that is made up of epithelial nests (e) that form the multilobulated parenchyma surrounded by loose connective tissue (stratum fibrosum internum, SI), and dense connective tissue (stratum fibrosum externum, SE) frame by the white line. The black line frames bundles of dense connective tissue attaching the JOO to the tissue surrounding it. Scale bar, 50 µm. (c) Detail of an epithelial nest where the nuclei of the peripheral epithelial cells are polarized towards the basement membrane (arrowheads). Scale bar, 100 µm. (d) Detail of the stratum fibrosum externum showing fibroblasts (arrowheads) and blood vessels (V). Scale bar, 100 µm. (e) Detail of the JOO showing nerve fibres (n) in the stratum fibrosum internum (SI) in relation to continuity between epithelial nests (arrows); white asterisk, vesicle with amorphous material; arrowheads, basement membrane. Scale bar, 20 µm. (f) Picrosirius red staining: the white line frames the stratum fibrosum externum (SE); the collagen of the SE is arranged in concentric layers and the collagen of the stratum fibrosum internum is arranged radially (arrowheads). Scale bar: 100 µm. (g) Immunoexpression of collagen I: the black line frames the SE (e, epithelial nest). Scale bar: 50 µm. (h) Immunoexpression of collagen type III was predominantly in the stratum fibrosum internum (SI); however, there is slight immunostaining in the stratum fibrosum externum. Scale bar: 50 µm

CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTIONS

Antonio José Mérida-García: Conceptualization; Formal analysis; Investigation; Writing-review & editing. **Jorge Murillo-González:** Funding acquisition; Investigation; Supervision; Writing-original draft; Writing-review & editing. **Elena Martínez-Sanz:** Formal analysis; Writing-original draft; Writing-review & editing. **Javier Catón:** Formal analysis; Investigation; Writing-original draft; Writing-review & editing. **Luis A. Arráez-Aybar:** Investigation; Writing-review & editing. **José Martín-Cruces:** Formal analysis; Writing-review & editing. **Teresa Cobo:** Formal analysis; Writing-review & editing. **José Antonio Vega:** Formal analysis; Supervision; Writing-review & editing. **José Ramón Mérida-Velasco:** Conceptualization; Formal analysis; Supervision; Writing-original draft; Writing-review & editing.

ETHICAL APPROVAL

All the procedures followed in this study were in accordance with the ethical standards of the Complutense University of Madrid (Spain) and in accord with the Helsinki Declaration.

PEER REVIEW

The peer review history for this article is available at https://publons.com/publon/10.1111/odi.13959.

DATA AVAILABILITY STATEMENT

The authors declare that all the data of this study are available on request from the corresponding author, Jorge Murillo-González.

ORCID

Jorge Murillo-González https://orcid.org/0000-0002-3455-2378
Elena Martínez-Sanz https://orcid.org/0000-0002-5492-8459

REFERENCES

Asgari, M., Latifi, N., Heris, H. K., Vali, H., & Mongeau, L. (2017). In vitro fibrillogenesis of tropocollagen type III in collagen type I affects its relative fibrillar topology and mechanics. *Scientific Reports*, *7*(1), 1392. https://doi.org/10.1038/s41598-017-01476-y

Bahcelioglu, M., Calguner, E., Erdogan, D., Elmas, C., Gozil, R., Keskil, S., & Kadioglu, D. (2005). The immunohistochemical approach to determine the origin and possible function of the juxtaoral organ in dogs. *Saudi Medical Journal*, *26*(10), 1529–1534.

Chievitz, J. H. (1885). Beiträge zur Entwicklungsgeschichte der Speicheldrüsen. *Archives of Anatomy and Physiology*, *9*, 401–436.

D'Andrea, V., Panarese, A., Di Matteo, F. M., Favoriti, P., Favella, L., Arcieri, S., & Filippini, A. (2015). Chievitz' juxtaparotid organ, free from cancer. *Annali Italiani De Chirurgia*, *86*, 503–507.

Gelse, K., Pöschl, E., & Aigner, T. (2003). Collagens–structure, function, and biosynthesis. *Advanced Drug Delivery Reviews*, *55*(12), 1531–1546. https://doi.org/10.1016/j.addr.2003.08.002

Ide, F., Mishima, K., & Saito, I. (2003). Melanin pigmentation in the juxtaoral organ of Chievitz. *Pathology International*, *53*(4), 262–263. https://doi.org/10.1046/j.1320-5463.2003.01466.x

Ide, F., Mishima, K., & Saito, I. (2004). Pacinian corpuscle in the juxtaoral organ of Chievitz. *Journal of Oral Pathology & Medicine*, *33*(7), 443–444. https://doi.org/10.1111/j.1600-0714.2004.00183.x

Kobayashi, Y., Nakashima, M., Wakakuri, T., Imaki, J., & Ito, M. (2015). Histology and immunohistochemistry of the developing juxtaoral organ in mice. *Annals of Anatomy*, *198*, 49–57. https://doi.org/10.1016/j.aanat.2014.11.004

Kuivaniemi, H., & Tromp, G. (2019). Type III collagen (COL3A1): Gene and protein structure, tissue distribution, and associated diseases. *Gene*, *707*, 151–171. https://doi.org/10.1016/j.gene.2019.05.003

Kusafuka, K., & Kameya, T. (2007). Juxtaoral organ of Chievitz, radiologically suspicious for invasion of lingual squamous cell carcinoma. *Pathology International*, *57*(11), 754–756. https://doi.org/10.1111/j.1440-1827.2007.02161.x

Mandl, L., Nerlich, A., Pankratz, H., & Hübner, G. (1993). The juxta-oral organ (Chievitz organ) – a sensory organ in the bucco-temporal area? *Der Pathologe*, *14*(4), 205–209.

Müller, S. (2016). Juxtaoral organ of Chievitz. In L. D. R. Thompson, & B. M. Wenig (Eds.), *Diagnostic pathology: Head and neck* (p. 377). Elsevier Health Sciences.

Pantanowitz, L., & Balogh, K. (2003). Significance of the juxtaoral organ (of Chievitz). *Head and Neck*, *25*(5), 400–405. https://doi.org/10.1002/hed.10209

Ricard-Blum, S. (2011). The collagen family. *Cold Spring Harbor Perspectives in Biology*, *3*(1), a004978. https://doi.org/10.1101/cshperspect.a004978

Soucy, P., Cimone, G., & Carpenter, B. (1990). An unusual intraoral mass in a child: The organ of Chievitz. *Journal of Pediatric Surgery*, *25*(11), 1200. https://doi.org/10.1016/0022-3468(90)90765-2

Suárez-Quintanilla, J., Martín-Cruces, J., Martín-Biedma, B., Cobo, J. L., Mérida-García, A. J., de Vicente, J. C., Junquera, L. M., Mérida-Velasco, J. R., & Vega, J. A. (2020). The juxta-oral organ of Chievitz (organum yuxtaorale) updated: Embryology, anatomy, function and pathology. *Annals of Anatomy*, *232*, 151582. https://doi.org/10.1016/j.aanat.2020.151582

Vadmal, M. S., Rossi, M. B., Teichberg, S., & Hajdu, S. I. (1998). Intraoral tumor of Chievitz in a child. *Pediatric and Developmental Pathology*, *1*(3), 230–233. https://doi.org/10.1007/s100249900031

Velasco, J. R., De La Cuadra Blanco, C., & Velasco, J. A. (2012). Development of the juxta-oral organ in rat embryo. *Anatomical Record*, *295*(5), 769–775. https://doi.org/10.1002/ar.22444

Zenker, W. (1953). Organon bucco-temporale (Chievitzsches Organ): Ein nervós-epitheliales Organ beim Mensch. *Anatomischer Anzeiger*, *100*, 257–265.

Zenker, W., & Salzer, G. M. (1961). Die Histologie des Chievitz'schen Organs. *Cells Tissues Organs*, *44*(4), 286–321. https://doi.org/10.1159/000141729

How to cite this article: Mérida-García, A. J., Murillo-González, J., Martínez-Sanz, E., Catón, J., Arráez-Aybar, L. A., Martín-Cruces, J., Cobo, T., Vega, J. A., & Mérida-Velasco, J. R. (2021). Functional hypothesis of the juxtaoral organ: Role of collagen types I and III. *Oral Diseases*, 00, 1–5. https://doi.org/10.1111/odi.13959