Limitations of the porcine model in post-total laryngectomy research

Emilien Chabrillac[1*], Thomas Hubert[2], and Agnès Dupret-Bories[3]

[1]Department of Surgery, Oncopole Claudius Regaud, University Cancer Institute of Toulouse – Oncopole, University of Toulouse, Toulouse, Occitanie, France; [2]Département Hospitalo-Universitaire de Recherche et d'Enseignement (DHURE), Lille University Hospital, Faculty of Medicine, Lille University, Lille, Hauts-de-France, France; [3]Department of Surgery, Toulouse University Hospital, University Cancer Institute of Toulouse – Oncopole, University of Toulouse, Toulouse, Occitanie, France

***Corresponding author:** Emilien Chabrillac, chabrillac.emilien@iuct-oncopole.fr

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Despite the advent of organ-preservation protocols, total laryngectomy (TL) remains a common procedure for locally advanced laryngeal and hypopharyngeal cancers. Since its first description in humans by Theodor Billroth 150 years ago, the surgical technique and perioperative care have been substantially refined with the help of various models. The porcine model is widely utilized in laryngeal surgery research[1,2] due to its close anatomical, dimensional, and histological similarities to the human larynx.[3] Furthermore, domestic pigs are inexpensive, readily available, and require less specialized husbandry than other large animal models.

For these reasons, researchers may attempt to perform TL in pigs or minipigs. However, a caveat should be noted. While the pig is an appropriate model for TL surgical training,[4,5] it does not seem suitable for post-TL research, including the study of pharyngocutaneous fistula,[6] which occurs within 15 days postoperatively.[7]

Our experience in performing porcine TL for post-TL research was unsuccessful. The experiment was conducted in the French National Veterinary School of Toulouse. Three male domestic piglets (GAEC Calvignac, France) weighing 45, 46, and 48.5 kg were delivered at 28 days of age. They underwent TL, with a gradual refinement of the post-operative care. Environmental conditions were controlled and appropriate, with heaters and adapted bedding to avoid aspiration of foreign bodies. Animals were single-housed. All animals wore a (cuffless) laryngectomy tube, tightly stitched to the skin. The first pig had a heat and moisture exchanger filter to help decrease the bronchogenic secretions. The two following animals had none, to allow them to expectorate sputum on their own and avoid trapping mucous plugs in the trachea. Post-operative monitoring included vital signs (temperature, respiratory rate, heart rate, oxygen saturation) and endotracheal suctions as required during the daytime. Endotracheal suctions did not retrieve a lot of sputum, and the animals showed no signs of chest infection. All three animals were found dead on post-operative day 2 ($n = 2$) and 3 ($n = 1$), without previous signs of breathing discomfort or deterioration of general condition. In all cases, crust formation or mucous plugging obstructed the tracheostomy tube or the trachea. Plugs were sometimes dry and never amenable to simple awake endotracheal suction; they would have required extraction under sedation. No accidental decannulation occurred.

Our findings are corroborated by the literature, as no cases of viable porcine TL have been published to date. The wearing of a tracheostomy tube in pigs has always been associated with several life-threatening challenges in the post-operative period. Only the team of Birchall *et al.*[8] succeeded in keeping the minipigs alive with a transplanted larynx and a tracheostomy for up to 14 days, attributed to complex monitoring and perioperative care; however, the animals still had a permeable larynx.[2,8] In another study by the same team,[2,8] transplanted minipigs were constantly monitored, including oxygen saturation, oxygen administration, and chest physiotherapy if needed. Crust formation was prevented by the constant wearing of a heat and moisture exchanger, cleaning of the tracheostomy tube, and hourly endotracheal suction with nebulization. Daily fibreoptic endoscopic airway examinations were performed to detect and manage crusting and chest infection. Despite all these precautions, two animals died of airway obstruction, one died of accidental decannulation, and only five out of 10 minipigs survived until the planned euthanasia at post-operative day 7.[1]

There is limited evidence in the scientific literature explaining these airway issues. In a computed tomography airway study of domestic pig airways, the trachea diameter was comparable to that of the human adult. However, the diameter, length, and branching angles of bronchi were noticeably different from those in humans.[9] The pig's trachea was also found to have similar biomechanical properties to those of humans in *ex vivo* studies.[10] However, the pig's anatomy has distinctive features. For instance, its cross-sectional shape is round rather than "C-shaped," as in humans. Indeed, in pigs, the trachealis muscle is covered by cartilage, rather

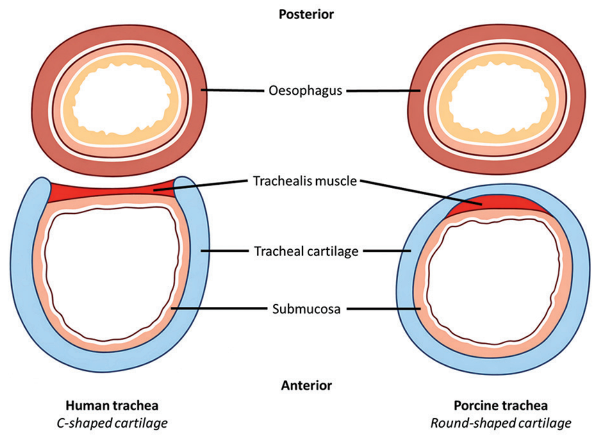

Figure 1. Schematic comparison of the cross-sectional view of the human trachea (left) and porcine trachea (right). Created with Paint 11.2601.441.0.

than connecting the ends of the tracheal cartilage rings as it does in humans[10] (**Figure 1**). The contraction of this smooth muscle decreases the diameter of the trachea, resulting in a collapse of the trachea, forming slits through which the air is forced, clearing tracheal content. Therefore, although this still remains to be proven, this muscle may have less of a narrowing effect in a more rigid trachea, with a round-shaped cartilage (**Figure 1**). Thus, it could be hypothesized that the pig's cough may be less efficient than that of humans. Furthermore, pigs are pronograde animals, with a horizontal tracheal orientation, and their cough is assisted by gravity when compared to orthograde animals such as humans. This is another potential explanation for a weaker physiological cough in pigs. However, this represents a mechanical disadvantage for airway clearance. Finally, the pig's thorax is narrower and shorter than that of humans, and, in veterinary practice, pigs are often considered to have a certain degree of cardiopulmonary weakness.[11]

Some authors have succeeded in maintaining Saanen goats, cats, and dogs alive after TL.[12-14] However, post-operative care was extensive, labor-intensive, and major complications occurred, including high mortality (11 of 19 goats died spontaneously within 3 months).[12] Of note, the tracheal anatomy of goats is variable but closer to that of humans, with either drop-shaped or C-shaped trachea on cross-sectional examination.[15]

Altogether, while we acknowledge that the porcine model is suitable for TL, researchers must be warned that it does not seem appropriate for post-TL research. Therefore, we encourage the development and publication of further large animal models.

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The authors declare no conflicts of interest.

Author contributions
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Ethics approval and consent to participate
The animal experiment referred to in this article received ethical approval by the local animal experimentation ethics committee #86 and was authorized by the French Ministry for Research (APAFIS#32524-2021072216434609). This animal experiment was performed in accordance with the ARRIVE guidelines and the French and European regulations on the protection of animals used for scientific purposes (EC Directive 2010/63/EU and French Decree 2013–118).

Consent for publication
Not applicable.

Availability of data
The data can be shared on a reasonable request to the corresponding author.

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