Appendix

Geometrical model

The CT-based anatomy that was imported into the 3D image-processing software Mimics (Materialise Software) included the entire nasal cavity (from the nasal planum and the frontal sinuses to the laryngopharynx) and the trachea (just proximal to the carina). After filling the cavities using manual reconstruction, as explained in the manuscript, the STL file generated was imported into computer-aided design commercial software (Rhinoceros, Robert McNeel and Associates). In this software, the nasal aperture and the trachea proximal to carina were cropped in relation to the first and last CT image, respectively. It is a requirement of numerical simulation that flow conditions at the external surfaces of the structure under study are set by the researcher (Hostnik et al., 2017). The geometrical model created for each dog was then exported in a readable format to generate the computational grids in the software (Ansys IcemCFD, Ansys).

Numerical method and boundary conditions

Flow conditions were set at the nasal and tracheal sections; zero flow velocity was set at the upper airway and tracheal walls. Atmospheric reference pressure was used in the simulations for computing all numerical variables. Additionally, flow was considered steady and incompressible (Craven et al., 2009; Hostnik et al., 2017). Canine inspiratory and expiratory flow was assumed to be steady (Craven et al., 2009). The steadiness of the flow is dependant on the Womersley number, W_o, defined as

$$W_o = D_h/2 \sqrt{(2\pi f / \nu)}$$

where W_o is a nondimensional number expressed as a function of the respiratory frequency f, the cinematic air viscosity ν, and the local hydraulic diameter D_h.

The Womersley number is an indicator of the unsteadiness of flow. When W_o is <1, the flow is assumed to be stationary, allowing steady-state modelling and solutions. A W_o >1 indicates the formation of nonstationary phenomena that require unsteady modelling and solutions. Craven et al. (2009) computed this number along the reconstructed nasal cavity of a Labrador retriever, assuming steadiness for inspiratory and expiratory flow and modelling flow as nonstationary for high-fidelity computations to simulate canine sniffing.

Our numerical simulations were performed using Ansys CFX software, which uses a finite-volume–based algorithm to simulate 3D airflow. The method used for solving the Navier-Stokes equations, which describe incompressible and turbulent airflow in canine airways, is iterative. Convergence of the numerical algorithm to solve the equations was considered achieved when the residuals of the error for momentum and continuity were $<10^{-4}$ (Craven et al., 2009).

Our simulations were carried out using a 16-node, Dual Nehalem (64 bits), 16-processor cluster with a clock speed of 2.33 GHz and 32 GB memory for each node. The first output was airflow velocity and pressure distributions in the canine upper airways. The spatial direction of the flow was generated using streamlines coloured to mark local velocity intensity at each location (Fig. 4); blue was the lowest velocity and red was the highest velocity. Airway pressure maps were generated using colour-coded overlays that represented the shape of the upper airway anatomy (Fig. 5). Different colours represented different pressure values, with blue being the lowest pressure and red the highest pressure.

Selection of the type of boundary conditions

In this study, we used flow conditions to determine pressure reduction and resistance. In particular, we used bodyweight-based inspiratory flow rates that were taken from the literature. Although breed-specific flows could provide more accurate results, because the dogs were sedated during the CT scan protocols, it was not possible to perform these measures. Breed-specific measures could be used in non-sedated animals. However, the flows used were adapted (and therefore individualized) to bodyweight, so that the results were not expected to differ considerably from previously published bodyweight-based inspiratory flow rates.

It is not possible to measure pressure inside the upper airways non-invasively because invasive devices that substantially affect airway measurements are required. In addition, since we were not interested in characterizing flow structure, which has previously reported (Craven et al., 2009), and the goal of our study was to estimate airway pressure reduction and resistance, we measured flow at the airway inlet and outlet in each dog. The flow at both extremities of the models, assuming mass conservation, could be used because the computational models were assumed to be non-compliant. This technique for computing pressure reduction has been used in human computational respiration physiology (Nowak et al., 2002; Brouns et al., 2007).

Mesh independence study

Prior to measurement of the canine upper airways, to establish the correct size for the numerical mesh, the effect of grid size was studied (Craven et al., 2009). The computational domain was progressively refined by increasing the number of tetrahedral elements dividing the volume of the canine airway to evaluate the accuracy of the numerical solution. We selected one dog (Labrador) that was meshed using a different number of elements. We increased the mesh size starting from a coarse grid of 10 million elements (Fig. 3). The finest grid contained roughly approximately 50 million computational cells.

The meshing procedure corresponds to the filling of the air passages using tetrahedral volumes in which the flow equations were computed. The dimensions of the tetrahedral edge length must be specified to generate the computational mesh. The tetrahedral element was selected for the discretization of the air passage volume because of the complexity of the canine upper airways studied (Hostnik et al. 2017, Craven et al. 2009).

For each grid, a numerical simulation was performed and the pressure used to compute the airflow resistance was evaluated (Fig. 3, Sections 1-4). The average pressure computed at each section was plotted as a function of the global number of elements (Fig. 3b). In agreement with Craven et al. (2009), the pressure computation was relatively insensitive to different mesh sizes (Fig. 3b). In particular, the relative error for a grid of approximately 20 million elements, using finer grids, was <5%. For this reason, and because of the huge increase in the computational costs of grid sizes with increasing elements, we used a mesh size of 20 million elements, which corresponds to a minimum edge length of approximately 0.2 mm.

Repeatability of the computational study

The repeatability of our computations was evaluated in one dog of each skull conformation (dolichocephalic, Great Dane; mesocephalic, Belgian shepherd; brachycephalic, English bulldog).

The geometrical reconstruction performed using the MIMICS software allowed clear definition of the black cavity representing the airways. A threshold automatically obtained using grey tones was used to highlighted black elements. In this way, it was possible to obtain a contour of the cavity in each CT image automatically; each contour was then filled manually. Because the procedure for obtaining the contour and location of the airway is provided automatically by the software, this procedure is repeatable.

The computational grids of the canine upper airways studied were regenerated using the same procedure and identical parameters. The computational grids were imported into the computational fluid dynamics (CFD) modelling package Ansys CFX, and the simulations for each dog were run a second time to assess differences between the two sets of results. In all cases, repeatability was >90%, confirming the agreement and reliability of the results reported using previously published methodology (Hostnik et al. 2017).

Method of comparison among groups

In this study, we could not perform any statistical analysis because of the heterogeneity of the dogs within each skull index group; the skull of each dog studied was geometrically and morphologically different. For this reason, results were unable to be compared statistically between skull index groups.

The aim of our study was not to identify anatomical relationships between the skull index groups using computed airflows or resistance, as performed previously in English bulldogs (Hostnik et al., 2017). We proposed a methodology based on medical images and CFD that could be used to quantify airway resistance non-invasively. Computational methodology was applied to canine upper airways to demonstrate varying resistance for different skull conformations. This methodology is widely used in human medicine, and its application has potential utility in veterinary species, as recently demonstrated (Hostnik et al., 2017) and could be especially important in the investigation of brachycephalic obstruction airway syndrome (Wykes, 1991; Dunié-Mérigot et al., 2010).