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THE ROLE OF HYOID BONE REPOSITIONING AND MANDIBULAR ADVANCEMENT IN MAINTAINING UPPER AIRWAY PATENCY

by
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ABSTRACT
OF THE THESIS OF

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Major: Orthodontics

Title: The role of the hyoid bone repositioning and mandibular advancement in maintaining upper airway patency

The hyoid is a mobile bone located at the base of the tongue. The mandible is the lower jawbone that hinges with the skull. Both bones are points of insertion of many upper airway (UA) muscles and are essential in maintaining UA patency. This is important as recurrent collapse of UA during sleep is characteristic of obstructive sleep apnea (OSA), a highly prevalent respiratory disorder with serious health risks. Indeed, OSA patients might have an inferiorly positioned hyoid and/or a retruded mandible. Surgical hyoid repositioning and/or mandibular advancement therapy(MAT) using a splint are viable options to treat OSA. However, MAT is only successful in ~50% of the cases. In fact, hyoid bone position may influence the outcome of MAT and may affect UA differently. Nevertheless, the precise influence of hyoid position and mandibular advancement altogether on UA function is unknown. The aim of this thesis is to determine the effect of hyoid position and mandibular advancement on UA patency.

Twelve anaesthetized, male, New Zealand White rabbits were tracheostomized and left to breath spontaneously via the caudal trachea (i.e. no UA airflow or muscle activity). The mandible was repositioned anteriorly from 0 to 4mm with 2mm increments, and along 70° vector directions using a customized mandibular advancement splint (MAS). Moreover, the hyoid was repositioned also from baseline within the mid-sagittal plane (anterior, cranial and caudal) from 0 to 4mm with 2mm increments, and along 45° vector directions using a customized device. The effect of hyoid and mandibular repositioning on upper airway function was quantified using the upper airway closing pressure Pclose (the negative pressure required to close the upper airway). Repeated measures three-way ANOVAs were performed to assess changes in key outcomes with hyoid and mandibular movements. The test of within-subjects was used to evaluate changes resulting from independent variables. The pairwise comparison was used to assess which pairs of conditions significantly differ from one another. Statistical significance for all the above analyses was inferred for P less than 0.05.

The interaction between mandibular advancement, hyoid repositioning direction and increment was statistically significant ($p<0.001$). The anterior movement of the hyoid and mandibular bones yielded the best outcome and the upper airway collapsibility decreased progressively when both bones were moved from 0 to 4mm. The more the hyoid and mandibular bones were advanced, the more P close was negative yielding a less collapsible upper airway. In contrast, when the hyoid bone was moved cranially or caudally, Pclose obtained was significantly higher at all incremental levels in comparison to Pclose obtained with the anterior component displacement. Mandibular

advancement of 4mm combined with ant-cranial 45° hyoid repositioning decreased upper airway collapsibility the most in comparison with other interventions.

This will be the first study to mechanistically examine the combined influence of mandibular advancement and hyoid bone repositioning on upper airway function. Final outcomes help understanding the role of the hyoid bone in MAT, and suggest potential combined therapy approaches to treat this serious health condition.

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LITTERATURE REVIEW

PART 1: THE UPPER AIRWAY

A. Structure and function

Anatomy of human airway is complex. It is involved in various functions such as swallowing, speech, and respiration. During phonation and deglutition, upper airway closure occurs. It is a compliant structure, comprising pharyngeal muscles that are anchored to the mobile hyoid bone. Upper airway is divided into 4 sections (Figure 1.1):

-nasopharynx: between the nostrils and the hard palate,

-velopharynx: between the hard and tip of the soft palate,

-oropharynx: from tip of the soft palate to the epiglottis,

-hypopharynx: from epiglottis to the glottis/larynx.

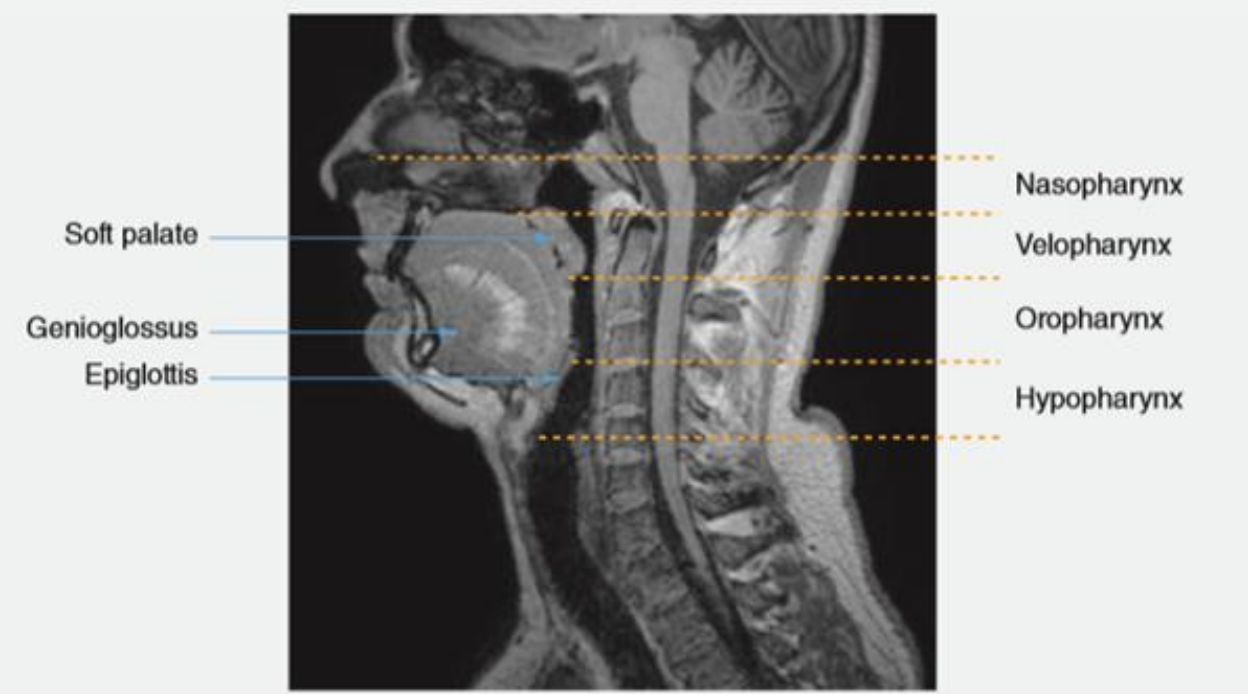


Figure 1 1:Schematic drawing of upper airway structure, Adapted from Carberry et al 2014 [1]

Upper airway anatomical assessment can be done using imaging methods: MRI (Magnetic resonance imaging), CT (computed tomography), cephalometric radiograph. Results obtained help in comparing upper airways in OSA and healthy patients. Characteristics of upper airways in OSA patients: smaller cross-sectional area (CSA), increased length, enlarged soft tissue including the tongue [2], fat accumulation, infero-posterior positioning of hyoid bone [3]. Upper airway is prolonged in males compared to females, justifying the higher susceptibility for males to develop OSA[1, 4].

B. Tissues

1. Bony structure

a. Maxilla-hard palate

i. Maxilla: (Figure 1.2) The maxilla is the principal bone of the midface. It is functionally and aesthetically important, defining facial architecture. It divides the nasal and oral cavities, shapes the upper jaw, and accommodates maxillary sinuses [5, 6]. A fusion exists between the right and left maxillary bones at the midline. The shape of the bone is pyramidal, its base is close to the nasal cavity, its apex being the zygomatic process and its body constituting the maxillary sinus[7] . It bridges surrounding facial structures through four processes: alveolar, frontal, zygomatic and palatine.

ii. Hard palate: (Figure 1.2) The hard palate incorporates 2/3 of the anterior part of the palate, the posterior part being the soft palate. It is a hard bony portion whose

fundamental structure consists of the palatine processes of the maxilla and the horizontal plates of the palatine bones.

It separates the oral from the nasal cavities, forming the floor of the nasal cavity and the roof of the oral cavity. The hard palate is enveloped superiorly by the respiratory and inferiorly by the oral mucosa. It has non-uniform transverse palatine folds called palatin rugae, that eases the motion of nutriments in the direction of the pharynx. It contains three foramina that serve as passage for neurovascular structures supplying the oral cavity: the incisive canal, the greater and the lesser palatine foramen.

b. Function of the hard palate:

- Creates a rigid floor to the nasal cavity preventing changes in pressure within the mouth from closing off nasal passages.
- Phonation, mastication and digestion aided by the presence of rugae [8-10]

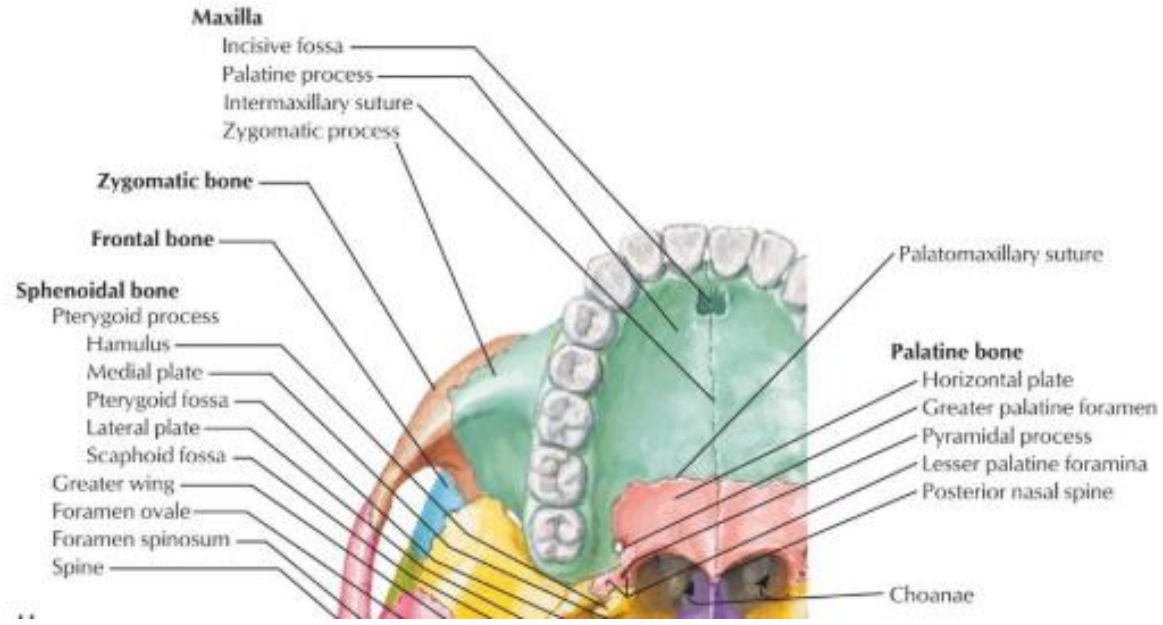


Figure 1 2:Schematic drawing of maxillary and palatal bone anatomy, Adapted from Atlas of Human Anatomy.

c. Mandible

The mandible is the broader bone in the cranium. It carries the mandibular teeth, it helps in mastication and shapes the jawline. It is positioned inferiorly relative to the maxilla and is formed by the body horizontally and the two rami vertically. Articulation with the temporal bone occurs at the level of the coronoid and condylar processes forming the temporomandibular joint which allows mobility [11].

Structure: Body of the mandible: it is the anterior part of the mandible, the body ends and the rami begin. The intersection between the body and the rami forms the gonial angle (Figure 1.3).

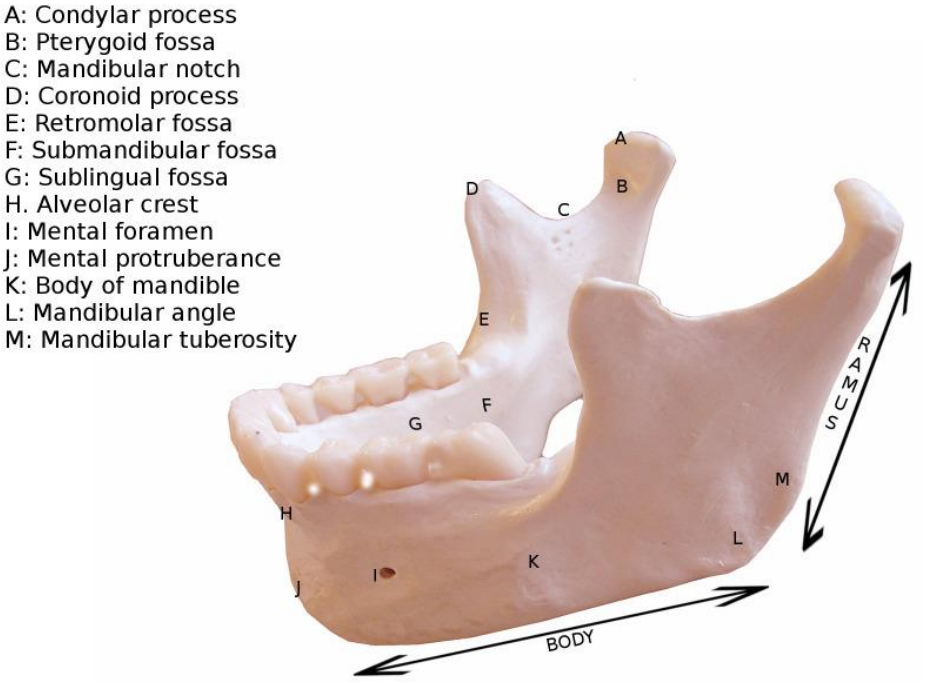


Figure 1 3: Schematic drawing of mandibular bone anatomy, Adapted from Anatomy, Head and Neck, Mandible. Grant Breeland; Aylin Aktar; Bhupendra C. Patel, 2020.

d. Hyoid bone

The hyoid bone is a horseshoe-shaped bone. It is located in the midline of the neck forming the base of the mandible anteriorly, at the 4th cervical vertebra posteriorly and above the thyroid cartilage.

Structure: The hyoid is composed of a body: median quadrilateral-shaped component, two greater and two lesser horns. It is considered as a freely floating bone [12]. Fibrous tissue attaches the horns to the body of the hyoid bone [13]. (Figure 1.4)

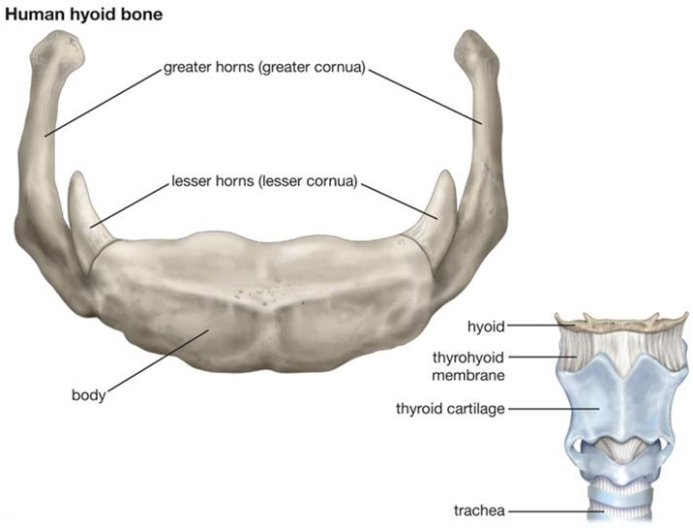


Figure 1 4: Schematic drawing of the hyoid bone and its attachment to the thyroid cartilage. The hyoid is a freely suspended bone. It is attached to cartilaginous and connective tissue, as well as muscles that control its position. Adapted from Encyclopaedia Britannica, 2010 [14].

2. Soft tissue structure (passive and active: muscles)

Upper airway is encircled by more than 20 muscles constricting and dilating upper airways.

Muscles are dived into four categories, balancing position of:

-soft palate: alai nasi, tensor and levator palatini,

-tongue: genioglossus, geniohyoid, hyoglossus, styloglossus

-hyoid bone: hyoglossus, genioglossus, digastric, geniohyoid, sternohyoid

-posterolateral pharyngeal walls: palatoglossus, pharyngeal constrictors [15]

Maintaining pharyngeal airway patency requires a synchronized activity of all muscles.

Muscles: (Figure 1.5)

The hyoid is as an anchor for supra and infrahyoid muscles: digastric, stylohyoid, mylohyoid, geniohyoid, sternohyoid, sternothyroid, omohyoid, and thyrohyoid [13]. It is suspended by the stylohyoid ligament from the temporal bone on right and left side. It is also attached to the thyroid cartilage by ligaments. In addition, the hyoid is bound to the cervical spine posteriorly along the cervical fascia [12].

Suprahyoid Muscles: total number of four muscles attaching the hyoid to the mandible, tongue, and skull. The digastric muscle has two bellies, posterior and anterior. The anterior belly derives from the digastric fossa of the mandible, while the posterior belly emanates from the mastoid notch. The stylohyoid muscle emerges from the temporal styloid process; and goes parallel to the posterior belly of the digastric. The mylohyoid muscle is a thin muscle that originates from the mylohyoid line of the

mandible; it sustains the floor of the mouth. The geniohyoid muscle is a narrow and short muscle that develops from the mental spine of the symphysis. All the above muscles attach to the body of the hyoid [12].

Infrahyoid Muscles: total of four pairs of muscles. The sternohyoid muscle arises from the manubrium, superior part of the sternum and attaches to the hyoid body. The sternothyroid muscle emerges from the back of the manubrium and is fixed to the thyroid cartilage. The omohyoid muscle is devised into a superior and an inferior belly. The superior belly of the omohyoid derives from the intermediate tendon of the omohyoid and inserts on the hyoid. As for the inferior belly, it emerges from the scapular superior border and inserts to the intermediate tendon of the omohyoid. The thyrohyoid muscle arises from the oblique line of the thyroid cartilage and inserts at the inferior aspect of the body of the hyoid and the greater horn [12, 16].

Other Muscles Related to the Hyoid: the hyoglossus and the middle pharyngeal constrictor

Elevation of the Hyoid: digastric, stylohyoid, mylohyoid and geniohyoid

Depression of the Hyoid: sternohyoid, sternothyroid, omohyoid, and thyrohyoid [17]

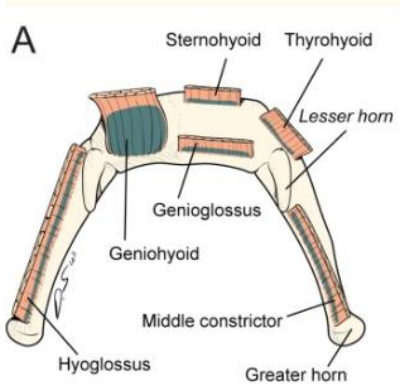


Figure 1 5: Schematic drawing of the hyoid bone and muscle attachements, Adapted from Gyun Ha et al, 2013.

The genioglossus is the biggest and most studied dilator muscle. It is responsible of protrusion of the tongue. It has a major role in keeping the upper airway patent. Its activity is reduced during sleep which leads to collapsibility of airway in some individuals [18]. In healthy subjects, the muscle contracts in an anterior direction during inspiration [18], this is not the case in OSA patients[19].

When awake, airway patency of OSA patients is sustained due to genioglossus muscle activity that is detected using electromyography activity (EMG). Nevertheless, when patients are asleep, a quick depletion in airway muscle tone occurs [20]. Consequently, individuals are more prone to airway constriction and collapse because of decreased muscular activity and anatomical susceptibility (e.g. smaller cross-sectional area (CSA), increased length, enlarged soft tissue including the tongue [2], fat accumulation, inferior positioning of hyoid bone [3]).

Through negative pressure, genioglossus muscle receives reflex from the receptors of upper airways. In fact, mechanoreceptors of upper airways are pressure-sensitive and are responsive to negative pressure. A reflex is delivered to genioglossus muscle to increase its activity and to counteract collapse. Even in healthy patients, reflex response of genioglossus muscle drops during transition to sleep, and both sleep stages: in REM and non REM sleep.

In human, sleep is composed of rapid eye movement (REM or “R”) sleep and non–rapid eye movement (NREM or “N”) sleep, with NREM further subdivided into various stages.[21]

Recent findings showed that during NREM sleep, genioglossus reflex is sustained when load increases in supine position [22, 23]. But reflex is decreased during REM sleep [22].

EMG reflex is similar between OSA and healthy patients during wakefulness [24].

C. Physiology and mechanical function

1. Upper airway geometry

Airway dimension is settled by bony structure of the craniofacial complex: mandible and hyoid bone anchoring muscles and soft tissue. The mean cross-sectional area (CSA) of upper airway in healthy patients was measured across multiple segments using several methods: acoustic reflection: 320-450 mm^2, fast CT: 59 mm^2 , conventional CT: 144 mm^2, 188 mm^2 and 138 mm^2 , MRI: 64 mm^2 [15]. Difference in quantification is due to: variation between patients, location of measurement, position of the patient: sitting or supine, and difference in imaging techniques. Measurements were settled when patient was in a wakeful state. Smallest diameter of the oropharynx is located at the level of the retropalatal area which is susceptible to constriction when patient is asleep.

2. Upper airway compliance

Compliance represents the functional radial stiffness of the upper airway. It is the ability with which airway can be deformed. In other words, it is the amount of upper airway lumen size variation (cross-sectional area or volume) in relationship to the change in intra-luminal pressure. Under the same applied load, a compliant tissue can deform more easily than another less compliant tissue. This measurement is a chief marker of possible collapse and a great indicator of OSA pathogenesis it gives an idea on the possibility of preservation of upper airway patency.

The drawback of using compliance to evaluate upper airway mechanics is that it is affected by neuromuscular reflexes [25].

3. Upper airway collapsibility

Collapsibility determines stability of upper airway and its resistance to closure. It is the likelihood of the pharyngeal lumen to narrow or close under a defined pressure. Collapsibility is useful to evaluate upper airway mechanics. Two prime measures characterize collapsibility: closing pressure (Pclose) and critical closing pressure (Pcrit).

Pclose : Pclose is evaluated under passive conditions, when upper airway muscles are inactive and airflow is absent through the upper airway. Upper airway narrowing occurs under negative pressure until reaching a threshold pressure where the airway totally collapses. The greater the negative Pclose value, the less collapsible the upper airway, and vice versa.

Human and animal studies have been undertaken to determine Pclose. Isono et al measured Pclose in humans (healthy and OSA patients) who underwent general anaesthesia and were mechanically ventilated. The upstream pressure was progressively decreased via a nasal mask and the upper airway surface variation with every pressure drop was recorded using videoendoscopy [26, 27]. A mathematical representation was then used to correlate pressure to area data and estimate Pclose. His results supported the anatomic hypothesis that apneics have a structurally narrowed and collapsible pharynx compared to healthy individuals [28]. However, animal studies are more advantageous because passive airways can be tested by excluding cofounding elements such as muscle activity and lung volume. Rabbit model was used, the trachea was sectioned and pressure was measured in a study done by Roberts et al. [29].The study

was initially performed on rabbits where the trachea was sectioned and the upstream/ downstream pressures monitored while the downstream pressure was decreased (Figure 1.6 and 1.7). The upstream pressure was decreased concomitantly with the downstream pressure in a patent upper airway. Once it was closed, the upstream pressure was not following the downstream pressure drop anymore. The downstream pressure at which this initially happened is the Pclose [29, 30].

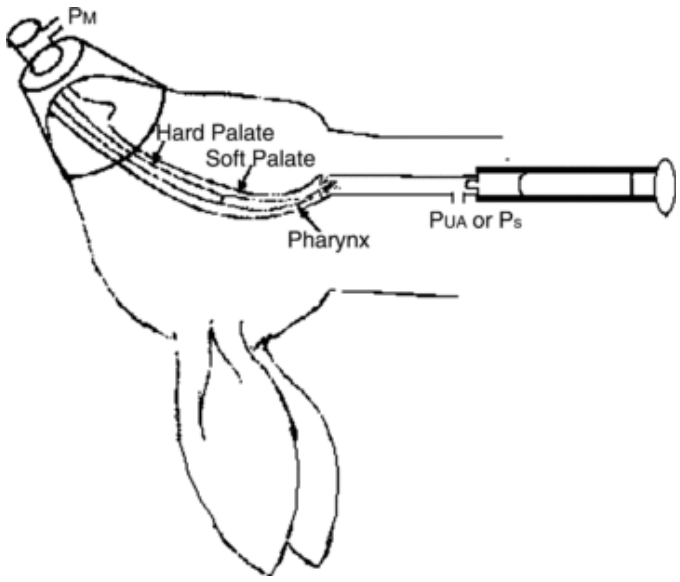


Figure 1 6: Schematic representation of the experimental set-up on rabbits. Closing and re-opening pressures were measured using a syringe to deflate/inflate the isolated upper airway whilst monitoring pressures in the mask (PM) and upper airway (PUA). Adapted from Lam et al. 2008 [29].

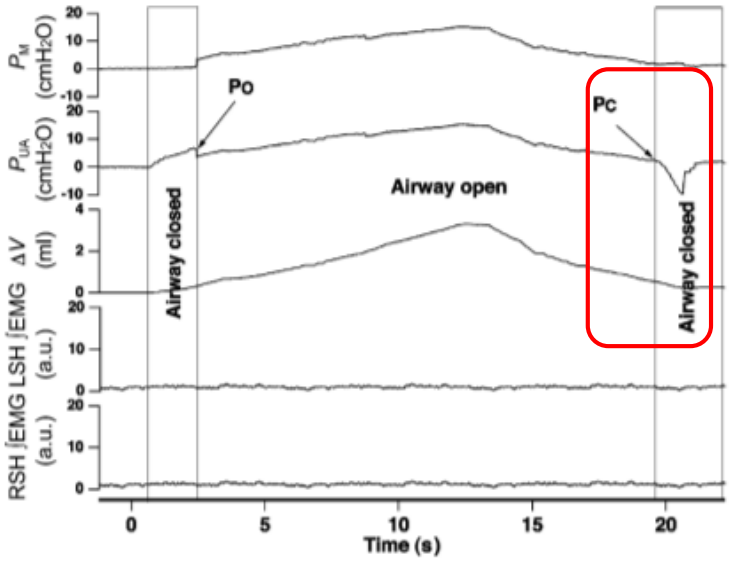


Figure 1 7: Data recordings of the pressure of upper airway closure Pclose in rabbits. The pressure in the upper airway (P_{UA}) and at the level of the mask (P_M) vary equally until the airway is closed then they become different. Pclose is then registered, Adapted from Kirkness et al, 2003 [31].

Pcrit: Pcrit is used frequently in human studies to determine upper airway collapsibility, especially during sleep [32-36]. It is reflective of the tissue pressure that leads to upper airway closure. In passive airways, it reflects pressure inside the airway at which airway collapses after flow limitation [37]. Pcrit is more positive in OSA individuals compared to healthy subjects. In general, a value of Pcrit above 0 cmH_2O indicates that the upper airway closure pressure is above atmospheric pressure and is associated with a compromised anatomy and obstructive apneas. A value below -5 cmH_2O refers to healthy individuals. Between 0 and -5 cmH_2O, the patient is borderline [38]. The more negative the value of Pcrit is, the less collapsible is the upper airway, i.e. it can withstand more negative intraluminal pressure before closing.

This technique helps in assessing decrease of collapsibility after certain therapeutic procedures: uvulopalatopharyngoplasty (UPPP), weight loss and to show the contrast between males and females: Pcrit is more positive in males compared to females [39, 40]. A Starling resistor model developed by Schwartz and colleagues led to the definition of Pcrit (Figure 1.8). This model comprises a collapsible tube (pharynx) interposed by two rigid segments (nose and larynx). It helps describing pressure and flow relationships in the upper airway [41].

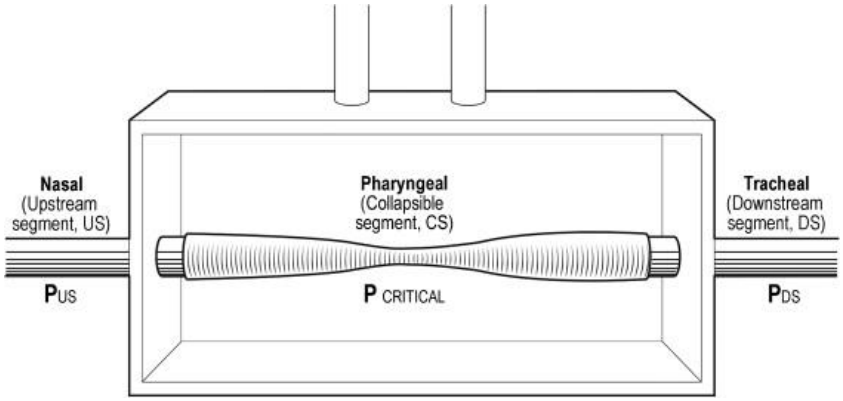


Figure 1 8: Schematic representation of the Starling resistor model of the pharyngeal airway. The pressure needed to keep the airway patent (Pcritical) is a function of the pressure upstream to the collapsing segment (PUS). Adapted from Hsia et al, 2015 [42]

Flow limitation occurs when airflow does not increase even with increased downstream inspiratory pressure. When this occurs, the upper airway is narrowed but not closed and the airflow is dependent on the pressure upstream to the constricted segment and the surrounding tissue pressure. Therefore, Pcrit is reflective of the tissue pressure that leads to upper airway closure [25, 43, 44].

Pcrit can be assessed passively or actively. Passive Pcrit is evaluated in patients that are anesthetized or asleep, where muscle activity is minimal. This is reflective of mechanical characteristics of upper airways. However, active Pcrit is evaluated when

upper airway muscles are functional reflecting active neuromuscular compensation for decreased intraluminal pressure. Active Pcrit measurements showed significant impairment in neuromuscular compensation in OSA patients [45].

4. Peripharyngeal Tissue Pressure

It is the extra-luminal tissue pressure (ETP) within the tissues surrounding the upper airway. Upper airway patency is in equilibrium between intraluminal and tissue pressure, thus the importance of ETP that should be taken into consideration. However, it is not feasible to measure peri-pharyngeal pressure in humans because it is an invasive procedure. Thus the importance of using an animal model: anaesthetized rabbit was initially used by Kairaitis and colleagues to determine ETP. ETP can be affected by head/neck position[46], mandibular advancement [47, 48], caudal tracheal traction [49, 50], changes in lung volume[51], sternohyoid muscle stimulation [52], and increased peri-pharyngeal tissue volume [53].

D. Upper airway physiology and OSA – traits contributing to OSA

OSA is a multifactorial syndrome. Contributing components comprise a narrowed or collapsible pharyngeal airway [54-56], decreased pharyngeal muscle activity throughout sleeping [24, 57-62], an increased loop gain[63-68], and a reduced arousal threshold during respiration[69, 70]. The corresponding contribution of these features differs considerably between subjects [71, 72].

Narrowed or collapsible pharyngeal airway: Upper airway size can drop due to enlargement of peri-pharyngeal tissues compressing upper airway walls. Moreover, airway collapse can be due to a retrognathic maxilla and mandible (jaw is posterior to

and behind where it should be when viewed from a lateral view) or because of a micrognathia (congenital abnormality of the jaws in which they are unusually small, particularly the mandible). Various imaging modalities (MRI and CT) have been used to demonstrate a reduced cross-sectional area of the upper airway in individuals with OSA [73, 74].

Poor anatomy: Moreover, poor anatomy of the upper airway bony or soft tissue structures is the prime suspect when aiming to determine the pathophysiology of OSA. The most observed anatomical feature is the altered position of the hyoid bone: inferiorly positioned. Upper airway dilator muscles are inserted on this bone and modification of its position might lead to muscle dysfunction in keeping the airway patent. Muscles can become less efficient in opening upper airway because of a modification in muscle's line of action and length/tension characteristics [45, 75]. Current studies show that the capacity of the pharyngeal dilator muscles to adequately compensate for increased mechanical loads during sleep is lessened in OSA patients compared to healthy individuals [76]. These anatomic and non-anatomic traits differ markedly between individuals; they can interact to cause or prevent OSA.

Decreased pharyngeal muscle activity during sleep and neuromuscular compensation:

During sleep, genioglossus muscle responsiveness is reduced in some OSA patients [18].

An increase in neural drive to upper airway dilator muscles is unable to occur as a reaction to negative collapsing pressure, thus contributing to OSA pathogenesis[72, 77, 78].

Increased loop Gain: The loop gain (LG) or ventilatory control system is defined as the ratio of ventilator corrective response to a disturbance. It modulates breathing via afferent feedback from chemoreceptors detecting carbon dioxide pressure. The instability of this system can cause OSA in some patients. For instance, high loop gain indicates excessive ventilatory reaction to a minor change in PaCO2; it is a large ventilatory response to a ventilatory disturbance. This leads to hypocapnia and therefore to a reduced stimulus to respiratory effort. Eventually, the upper airway collapses in some individuals as a result to the high loop gain, which is the case in ~30% of OSA patients [34]. With a similar Pcrit, loop gain is prone to be elevated in patients with OSA relative to control patients [79].

Reduced respiratory arousal threshold: A reduced arousal threshold contributes to the development of the pathology: it means that when patients are faced with an increased respiratory demand, they would wake up easily. A reduced threshold was observed in greater than 33.3% of OSA patients. These individuals have a disrupted sleep continuity, they wake up prematurely and no opportunity is given for upper airway muscles to restore airflow during sleep [70, 77, 80, 81]. It may be a therapeutic target; nevertheless, invasive procedures are required to do the measurements: pressure catheter at the level of the epiglottis or the oesophagus [82]. Some patients have a high respiratory arousal threshold. This increased threshold gives more time for a counterbalance of poor upper airway anatomy by recruiting pharyngeal dilator muscles [83].

E. Factors Influencing Upper Airway Tissue Mechanical Properties

Various factors have an impact on mechanical properties of upper airway tissues such as: fat deposit, tracheal traction, posture and head position along with nocturnal rostral shift of fluid.

Fat content can narrow upper airways and can lead to an increase in Pcrit and risk of OSA. This induces alteration in passive tissue stiffness [84]. Fat accumulation in upper airway region differs between males and females affecting collapsibility of upper airways and prevalence of OSA differently [85].

Caudal tracheal traction augments stiffness of the airway walls following inspiration and increase in lung volume. In rabbits, it leads to a reduction in collapsibility of upper airways [49, 86], caudal displacement of the hyoid bone and increase in size of upper airway following tracheal traction [124]. However, decreased lung volume increases severity of OSA [87]. In fact, when lung volume rises, upper airway wall tissues are subject to elongation leading to an increase in stiffness and the opposite occurs when lung volume drops [50, 88]. Males are prone to have more collapsible upper airways relative to females due to a decrease in lung volume and increased obesity rate [40, 89].

Posture affects mechanics of upper airways specially when patient is sleeping due to the supine position and backward movement of the tongue leading to airway closure [90, 91]. One of the treatment options for some OSA patients is to sleep in the lateral position to decrease apneas and hypopneas during nighttime [91].

Head position: can also affect upper airway collapsibility. Head extension decreases airway collapsibility while flexion increases it [37, 87], this can also modify the location of collapse [87]. However, collapsibility was not affected by head rotation [87]. These

differences have been attributed to tracheal traction, although Isono and colleagues [37] suggest that the compliance of the airway walls decreased with neck extension due to increases in airway cross-sectional area.

Jaw position: may also influence passive collapsibility. Collapsibility increases during mouth opening because of a constriction at the level of the oropharynx [37]. It also increases with age [24] due to an increase in obesity and airway resistance, so this appears likely to be related to airway narrowing rather than specific age-related changes in tissue mechanics. In addition, advancement of the mandible, neck extension, and lateral position decreases closing pressure in subjects under general anaesthesia [28]. Nocturnal rostral shift of fluid increases upper airway collapsibility [18, 95], due to increase in mucosal edema that narrows the airway and softens surface layer on the pharyngeal wall [25]. The contribution to OSA pathophysiology are not clear yet.

F. Models of Upper Airway Behavior

Biological process can be acknowledged by using a simplified representative model. These systems are used to study a concept or to test a hypothesis. However, a unique model cannot represent biological, mechanical and physiological characteristics. Various models characterize upper airways including conceptual, physiological, physical, mathematical and computational models. The physiological model includes both human and animal subjects. Each of these representation can have benefits as well as drawbacks.

Conceptual model: Initially developed by Remmers et al. to examine equilibrium of forces between intra-luminal pressures and forces aiming to open the airway [92] . At a later stage, this concept was developed more by Isono et al, they included an

evaluation of the passive mechanical properties of the pharynx by relying on the balance of pressures [26]. Moreover, a third major conceptual model is the Starling resistor model (Figure 1.8) that simulated the upper airway to a collapsible tube fixed at both ends and floppy in the middle segment. The tube is surrounded by a chamber filled with air to represent peri-pharyngeal tissue pressure. However, conceptual model helps to interpret the fundamental function of upper airway. Anatomical and physiological concepts cannot be understood using this model.

Physiological models: using human or animal model.

Human model: using a conservative and non-invasive approach not to cause damages or injuries to the patient. Undergoing unnecessary intervention on humans is unethical. This limits human studies and makes them a non-viable option. Recently, there has been a focus on studying response to OSA treatments, such as mandibular advancement and CPAP therapy through human studies. [36, 93, 94].

Physical, mathematical and computational models: These models are advantageous because they give the opportunity to control various parameters while eliminating biological disparity. This provides a better understanding of precise mechanisms and complex procedures. In addition, they allow the performance of strategies that are impossible to achieve physiologically. The computational finite element modelling approach was used in the study done by D. Salman and J. Amatoury 2021 to examine the role of hyoid bone position in upper airway patency and tissue mechanics.

A simplified representation of this model is the use of Starling resistor. This model relates airflow, intraluminal pressure and peripharyngeal tissue pressure [95]. It helps measuring airflow pressure through the system by finding the difference between the

pressure upstream and Pcrit and is independent of the pressure downstream[96]. However, the major drawback of the Starling resistor is that it excludes muscular activity and functional anatomy of the airway walls. [95].

Animal models: This model has various benefits; it is used when the procedure requires the sacrifice of the subject or application of invasive techniques. Another advantage of using an animal model is that sample size can be reduced in comparison to human studies due to the decreased variability between animals belonging to the same species. However, this model does not deliver accurate outcomes that are applicable to humans due to critical anatomical and physiological differences. Moreover, animal models are financially and technically difficult to manipulate. They require a high budget and some procedures are difficult to achieve. The rabbit is a good model to study upper airway mechanics and physiology. It was previously used in several studies [47, 97-100].

The Anaesthetized Rabbit model and upper airway physiology

Upper airway physiology has been studied using numerous animal species including mice, rats, dogs, cats and rabbits. Studies included tongue displacement and upper airway vibration[47], initiation of hypoxia and/or hypercapnia and implementation of tracheal traction[49], stimulation of upper airway muscles [29, 52, 101, 102] or nerves, including the hypoglossal nerve [103] as well as mandibular advancement [48, 104, 105].

To evaluate sleep breathing disorders, animal models were assessed. In some animals, such as in bulldogs and female pig in China, OSA was present without the need to induce it. For instance, English bulldogs had the syndrome due to a wide soft palate and a constricted oropharynx. These findings were elaborated by Hendricks and colleagues

in 1987. Moreover, a female pig in China was found to have an atypical narrow upper airway anatomy [97, 106]. However, these animals are not extensively reachable for research. Induced OSA models were necessary, and so features of OSA were developed in other species such as cats, rabbits and other breeds of dogs. Invasive and non-invasive procedures were applied. Invasive approaches included tracheotomy, administration of silicone injection at the tongue of a rabbit to impede breathing [97, 106]). Non-invasive techniques involved hypoxemia for few months allowing further investigations of chronic sequelae that are comparable to human diseases [97, 106].

Upper airway and respiratory physiology are studied based on the rabbit model. The most important aspect of use of this model is its similarity to the human model, making it the ideal model to be used for upper airway investigations. Particularly, unlike most non-primates, the hyoid bone is mobile in the rabbit as in human [48, 107].

In fact, the hyoid is connected to the thyroid cartilage through the thyrohyoid membrane similarly to the human anatomy (Figure 1.9). Moreover, upper airway anatomy and sections are similar to that in humans: nasopharynx, velopharynx and hypopharynx (Figure 1.10). However, the velopharynx is longer in rabbits compared to humans due to the long soft palate, which extends beyond the epiglottis [108]. Hence, the rabbit oropharynx is not involved in breathing as in the human (i.e. it is bounded by the soft palate posteriorly).

In addition, the rabbit's upper airway has similar soft and hard tissue constituents as humans and the airway is susceptible to collapse [86, 97]. Furthermore, neurological and innervation similarity do exit specially for the cranial nerve XII. [109].

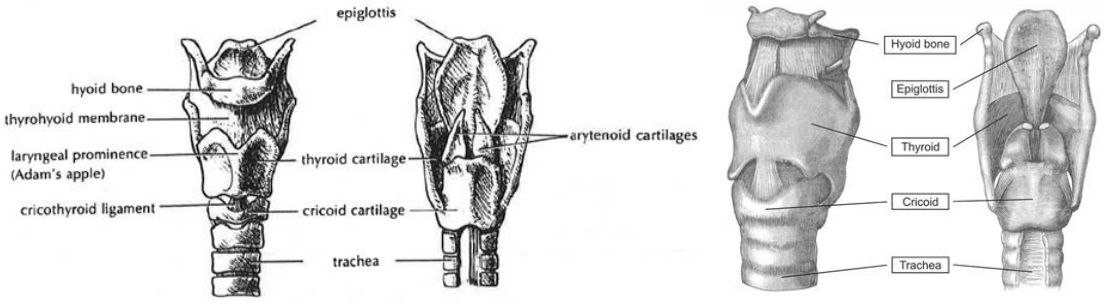


Figure 1 9: Schematic representation of the similarity between the anatomy of the larynx in humans and in rabbits. on the left: scheme of the rabbit's larynx anteriorly (on the left) and posteriorly (on the right). Adapted from Wingerd, 1985 [108]. On the right: scheme of the human larynx anteriorly (on the left) and posteriorly (on the right), Adapted and modified from Kniesburges et al, 2011 [110]

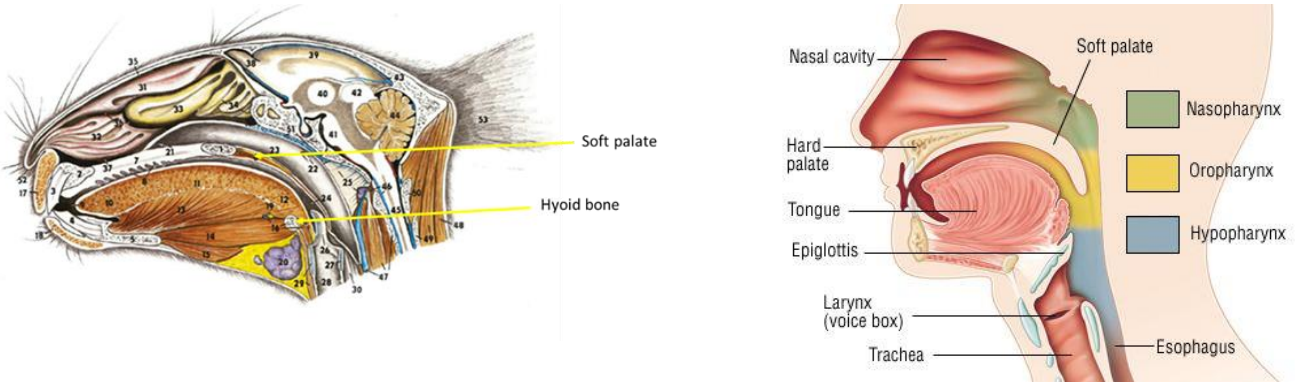


Figure 1 10: on the left: scheme of the rabbit's upper airway anatomy. Adapted from Bioscience Oxford University Journal,2017. On the right: scheme of the human upper airway anatomy. Adapted and modified from A. Zaghawa et al,2019 [111].

Summary

The upper airway is a collapsible structure but maintaining its patency is a necessity for breathing. Several factors such as the negative pressure during inspiration and the extra luminal tissue pressure will promote its closure. In contrast, other dilating forces such as the activity of the dilator muscles and the tracheal traction from the lungs expansion during inspiration will ensure its patency. The equilibrium of the dilating and collapsing

forces is a necessity and the loss of this balance will lead to breathing disorders such as obstructive sleep apnea (OSA).

PART 2: OBSTRUCTIVE SLEEP APNEA

A. Definition and epidemiology

Obstructive sleep apnea (OSA) is a condition characterized by repeated episodes of partial or complete obstruction of the respiratory passages during sleep [112]. It is a common disorder with significant health impact [113] . It affects at least 6-17% of middle to older aged adults [114, 115]. It is more prevalent in males compared to females: 9% in women and 24% in men [116]. The prevalence of this disorder is increasing over time [117]: in the US population OSA increased by 14-55% between 1988–1994 and 2007–2010 [116] . Diagnosis and treatment of OSA is crucial to perverse patient's health.

B. Risk Factors

Main risk factors for OSA are male gender, older age, obesity (increased BMI) and central fat distribution [116] [118].

Male gender – OSA is 2-3 times more prevalent in males compared to females. However, the risk is analogous in postmenopausal women [119-122]

Older age – OSA boosts from young adulthood to the age of 60-70, then become stable with no further increase[119, 123, 124]

Obesity – OSA is interrelated with the body mass index(BMI) and fat distribution[114, 120]. Risk of OSA increases by six-folds with weight gain of 10% [125]. A study was conducted to examine the correlation between OSA severity and BMI. The higher the BMI, the more severe is OSA in males as well in female patients[119].

Risk factors for OSA can be anatomical: craniofacial abnormalities, upper airway deformities such as: hyoid bone positioned inferiorly, narrower pharyngeal airway, larger volume of lateral pharyngeal wall and soft tissue surrounding upper airway [116]. Short mandible, abnormal maxilla, broad craniofacial base, and the common feature seen in children: hypertrophy of tonsils and adenoids.

Less probable risk factors:

Smoking – OSA is 3 times more prevalent in smokers compared to nonsmokers or patients who had quit smoking.[126]

History of OSA or snoring in the family –Genetic, environmental and behavioral factors are present among individuals belonging to the same family. The genetic factor can be related to the craniofacial structure[127].

Others– Prevalence of OSA was double in patients having nasal congestion relative to controls [128]. Medications: benzodiazepines, narcotics or alcohol intake can aggravate OSA. The causal relationship is questionable [129].

OSA has significant health impacts [113]: it causes episodic hypoxemia, nocturnal sympathetic nervous system activation, elevates blood pressure and markers of oxidative stress, inflammation, and hypercoagulation. Population-based and sleep-clinic–based cohort studies have shown an association between obstructive sleep apnea and cardiovascular events, particularly stroke [130]. Multiple studies demonstrated a 2-fold increased risk of stroke in patients having OSA compared to patients without OSA[131, 132]. OSA increases also the risk of obesity due to inactivity and neurohormonal dysregulation[116]. (Figure 2.1)

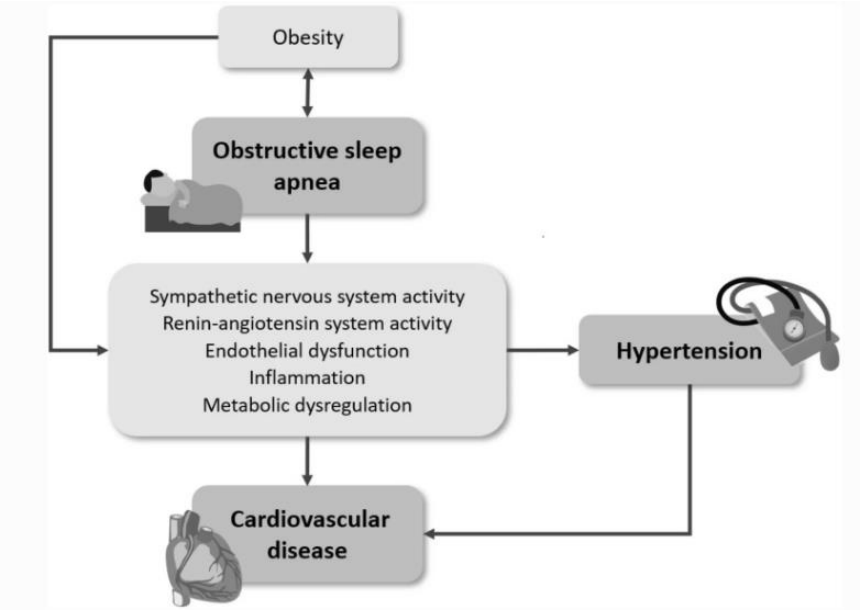


Figure 2 1: Schematic representation of the pathophysiology of Obstructive Sleep Apnea: Hypertension, and Cardiovascular Risk. Adapted from Salman et al. [116]

1. Pathophysiology:

Apneic episodes are recurrent in OSA patients leading to intermittent hypoxia. This induces a high arterial pressure and cerebral vascular resistance [116]. A cohort study, including 6132 participants, showed that the prevalence of hypertension increased with the severity of OSA. Prevalence of hypertension was: 59% in mild, 62% in moderate, and 67% severe sleep apnea patients [133]. Blood pressure increases in a linear direction with increased severity of OSA [134].

C. Diagnosis (PSG, AHI)

Those who have OSA may complain of tiredness, excessive day-time sleepiness, insomnia, or morning headaches, but many are asymptomatic [112]. Patients report

intense snoring, disturbance or discontinuation of breath while sleeping. This can be identified during the questionnaire of screening for preoperative care [135].

1. Questionnaire

Daytime sleepiness – Prevalent characteristics of OSA. Patients cannot persist fully attentive and awake. They can easily fall asleep during the day. Epworth Sleepiness Scale (ESS) is used to quantify patient's sleepiness and fatigue [136, 137] . An ESS score exceeding 9 demonstrates unusual sleepiness and requires additional testing. Fatigue Severity Scale (FSS) is also helpful in identifying the problem [138, 139] .

Snoring, choking, gasping during sleep – Apneic events, silence followed by intense snoring are prevalent characteristics of OSA [140].

Morning headaches – Disclosed by 10-30% of patients with untreated OSA [141]. The reason is not well-known, it is multifactorial and complex; probable causes: elevated carbon dioxide levels in the blood known as hypercapnia, vasodilation, high intracranial pressure, and defective sleep condition [142].

2. Physical examination

Obesity – A high BMI of at least 30 kg/m^2: is a frequent clinical discovery in OSA patients. [143]

Constricted airways at the level of the oropharynx– due to a smaller or retrognathic lower jaw, a larger tongue, enlarged tonsil, an oversized uvula, a constricted palate, deviation of the nasal septum, and polyps in the nasal cavity [144].

Signs of correlated conditions–hypertension and heart failure [143].

The gold standard for diagnosing OSA: is polysomnography (PSG) [116, 145]. During PSG, sensors are used to test sleep-breathing disorder. Airflow is calculated at the level

of the nasal and oral cavity using sensors. Snoring level and intensity is also registered. Oxygen saturation in the blood and carbon dioxide (CO_2) are measured. Electroencephalography (EEG), electrocardiography (ECG), and body position assessment are also included [135] . Severity of OSA is admeasured using the apnea-hypopnea index (AHI, events/hour) [116] [134]. Three OSA grading scale do exist: mild (5-15 episodes/hour), moderate (15-30 episodes/hour), severe (>30 episodes/hour)[145-147]. A diagnosis of OSA is based on: history, apnea hypopnea index (AHI) ≥ 5 and respiratory disturbances on PSG [145].

D. Treatment options

Various treatment options are available to treat obstructive sleep apnea. Continuous positive airway pressure (CPAP), oral appliances, surgery, hypoglossal nerve stimulation and conservative approaches including weight loss and positional therapy [148].

1. Continuous positive airway pressure (CPAP)

First-line treatment for OSA is: continuous positive airway pressure (CPAP) (Figure 2.2), highly efficacious in reducing sleep-disordered breathing events [149].

Figure 2 2: Schematic drawing of CPAP device. Adapted from American Association of Sleep Technologists *(*AAST*).*

Indication: CPAP treatment is indicated for patients having moderate to severe OSA and for mild cases having underlying cardiovascular problems [150, 151].

Treatment outcome: Use of CPAP decreases apnea-hypopnea index (AHI). Thus reducing symptoms, enhancing quality of life, and avoiding life-threatening cardiovascular incidents [149]. It lessens daytime sleepiness and improves neurocognitive function [152]. Cardiovascular incidents decreased in patients wearing CPAP for at least 4 hours every night [130].

However, about half of all patients with OSA who try CPAP therapy are either completely intolerant or only partially adherent [117]. Compliance was defined as use of CPAP for at least 4 hours/night for 70% of the nights. Patients reported compliance ranging between 65%-90%. Patients usually overestimate CPAP use.

Adverse effect: Common problems leading to lack of adherence with CPAP are claustrophobia, nasal congestion, skin irritation, leakage due to an ill-fitting mask[152].

Patients education can also play an important role[153]. Patients that are more educated and that are frequently followed by healthcare provider, are more compliant to CPAP therapy [150, 151]. Addition of heated humidification can also increase compliance[152]. Due to different existing problems, 50% of patients fail to adhere to CPAP therapy[117]. Accordingly, alternative treatments for OSA are clearly required [79].

2. Mandibular advancement splint (MAS)

MAS can be used after failure of CPAP as a substitute or a second-line treatment [83].

Indication: First choice to treat primary snoring without obstructive sleep apnea [154], mild-to-moderate OSA patients or CPAP-intolerant severe OSA patients[154, 155]. The strong predictors of success with MAS include: mild OSA of non-obese young females [156, 157] with retruded mandible and a smaller neck circumference and oropharynx [158], a short soft palate and long maxilla . A polysomnography test can be done to evaluate success[154].

Treatment outcome: It protrudes the mandible, widens the oropharyngeal and stretches the muscle of the tongue. Prevents collapse of the tongue against the pharyngeal wall. It is effective in diminishing snoring and obstructive events [159, 160]. It expands the oropharynx and velopharynx by moving the soft palate and the tongue more forward (Figure 2.3).

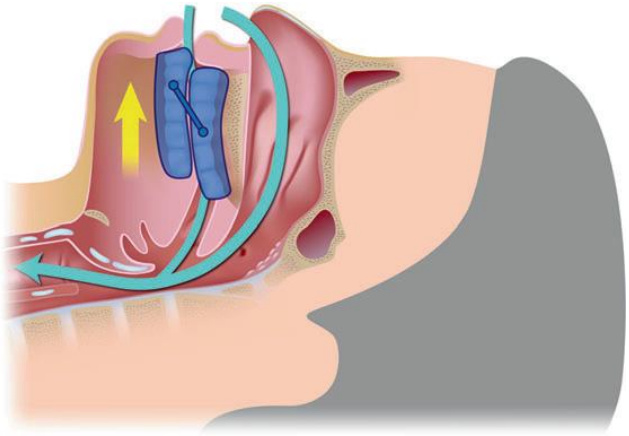


Figure 2 3: Schematic drawing of the effect of the MAD as it advances the mandible and opens the posterior airway space. Adapted from Contemporary Oral Medicine book.

Adherence rates are reported to be superior than those for CPAP by almost one and a half hour per night. Health benefits are identical to CPAP [83]. Mandibular advancement decreases collapsibility of the pharynx with variability between subjects. MAS decreases OSA via enhancement of pharyngeal calibre and improvement in airflow [161]. But oral appliances are not as efficient as CPAP in reducing AHI. Only 42% of oral-appliance patients have AHI of less than 5 events per hour after use of the device[151, 162].

MAS designs and treatment stages: Using MAS necessitates an interdisciplinary approach starting with the assessment of a physician, followed by consulting with a dentist. The dentist will take impressions of the teeth and registration of the bite to fabricate a customized device. Different MAS designs and features are commercially available. It can be a one-piece appliance (monobloc) or two-piece design that is composed of two separate upper and lower parts. The two-piece appliances allow a degree of lateral jaw movement and variable amounts of mandibular advancement [162,

163]. There are no defined parameters based on scientific evidence determining the amount of advancement required to achieve an optimal result with the MAD. However, titrating the mandibular advancement has an important influence on treatment efficacy [98]. Titration is done gradually by advancing the mandible forward. Kato et al reported that 2-mm of mandibular advancement improved oxygen desaturation index by 20% [164]. At least 50% of maximum protrusion is desirable to have a positive therapeutic outcome[165-167]. Monthly visits are required for the first five months. Dentist should check for compliance and appliance wear, dental occlusion, and potential side effects: on teeth, soft tissue, and joint. Evaluation of efficacy and comfort reported by the patient is also mandatory. MAS can be used for 3 years on average before needing replacement. Fracture, discoloration, and absence of retention are indicators for urge of replacement [147].

Side effects: Mucosal dryness (86%), tooth discomfort (59%), hypersalivation (55%) and dental changes [168]. Not all patients are eligible for oral appliance therapy due to dental exclusions such as insufficient teeth and periodontal disease. To retain MAS, eight to ten healthy teeth should be available in the mandibular jaw, the maxillary jaw can be edentulous [147]. Treatment adherence can be reduced by unwanted dental changes, temporal mandibular pain, and excess salivation [83]. Recent studies suggest that the presence of temporomandibular disorder (TMD) should not be a contraindication for the use of advancement device [146, 147]. If the patient is suffering from TMD problems, the dentist has to treat the joint before starting splint therapy. The dentist's role is of high priority in managing MAS therapy, as it is a life-long treatment and is associated with dental and joint side effects [146]. MAS is not effective in all patients. It is completely effective in only 36% to 70% of OSA cases[147]. It is

beneficial in 50% of patients on average [163]. Success rate depends on the sort of device used, seriousness of OSA, amount of protrusion obtained and body mass index. MAS therapy is less advantageous in patients having severe OSA with an AHI > 30 events/hour, and in patients having a high BMI [162].

MAS versus CPAP: MAS has been compared against CPAP, as a current standard of care, in many randomized trials. In moderate to severe cases, MAS was less effective in reducing AHI [94]. Sleep tests show that AHI levels were reduced to normal when treated with CPAP but not with MAS [31]. However, patient's compliance was better with MAS. Improvements in terms of daytime sleepiness, quality of life, neurocognitive performance, and blood pressure were comparable in both treatments. Therefore, although treatment with MAS resulted in less AHI improvements, this does not reflect on the obtained health benefits [41]. Compliance with MAS is the main factor that compensates for the superiority of the CPAP in lowering the AHI value. Greater usage may counterbalance the lower treatment efficacy and result in overall equivalent treatment success [42]. MAS has been reported to be worn for a longer period of sleep time compared to CPAP [28]. Ultimately long-term studies of the comparative effectiveness CPAP and MAS are needed to provide robust evidence that supports substituting CPAP with MAS.

3. Muscle functional therapy: Hypoglossal nerve stimulation (HNS)

Patients who are intolerant to CPAP may use HNS as an alternative. Patients complied by 86% at 1 year compared to 40%–60% compliance with CPAP [169-171],[172]. HNS consists of an implantable hypoglossal nerve stimulator (Figure 2.4).

These devices utilize neuromodulation to dilate/reinforce the airway and reduce side effects associated with traditional surgery [113].

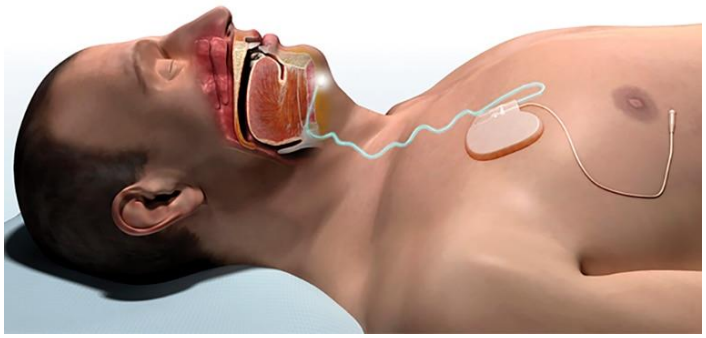


Figure 2 4: Schematic drawing of muscle functional therapy: Hypoglossal nerve stimulation. Adapted from Mayo Clinic.

Treatment outcome: It activates the largest upper airway dilator muscle: the genioglossus, to increase airway patency, but this too has variable efficacy [173, 174].

Indication: HNS can be done in patient with a BMI <35 kg/m^2, AHI: 20-50 events/hour [175].

Adverse effects: pain (6.2%), tongue abrasion (11%), device malfunction (8.8%)[113]. It can also induce paresthesia, salivary flow changes and lip weakness [176].

4. Surgical interventions (UPP, MMA)

Surgical treatment manifests excellent clinical result in short-term symptoms and long-term health. However, surgery does not cure OSA. This therapeutic option for OSA remains controversial. Assessing surgical therapy is complex since placebo control and randomization are not practical with invasive procedures. However, controlled

studies pointed out important profit of surgery and combinations of surgical procedures [177]. To increase airways patency, different surgeries are available. Procedures comprise: uvulopalatopharyngoplasty (UPPP), tracheostomy, genioglossus advancement, glossectomy, hyoid suspension, maxillomandibular advancement (MMA), and combinations of the various interventions[152].

➔ Uvulopalatopharyngoplasty (UPPP):

Procedure: excision of the soft palate and uvula (Figure 2.5), with CO2 laser operation. An increased amount of side effects led to a drop in UPPP surgeries over the last 10 years [178]. It was one of the most common procedure executed for sleep apnea due to the fact that the retropalatal area is frequently obstructed. Additional surgeries target collapse in the nasal or retrolingual regions. These interventions can be performed simultaneously with UPPP such as: tonsillectomy, septoplasty, turbinate procedure, mandible/genioglossus advancement [179, 180].

Figure 2 5: Schematic drawing of uvulopalatopharyngoplasty. Adapted from Sleep Apnea Surgery Barrington IL, Specialty Care Institute.

Side effects: at 6 month-post surgery: 8% swallowing difficulties, 6% had voice issues, 4% had increased mucus production, 2% had gag reflex, 2% had dry throat, 2% had periodic tonsillitis. At 24-month follow-up, patients reporting presence of side effects decreased from 38 to 31%. But side effects were still present [179, 180].

➔ Maxillomandibular advancement (MMA): (Figure 2.6)

Maxillomandibular advancement (MMA) surgery, to enlarge the pharyngeal space by expanding the skeletal boundaries of the maxilla and mandible, is currently considered the most efficacious surgical procedure for treatment of OSA, particularly severe OSA[163].

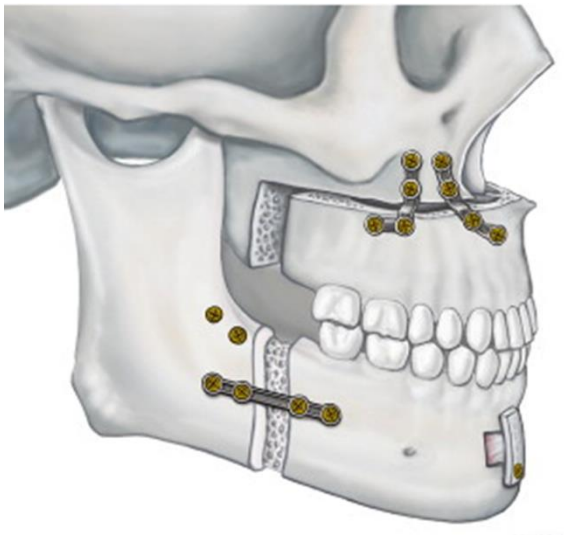


Figure 2 6: Schematic drawing of MMA surgery. Adapted from Skeletal Surgery for Obstructive Sleep Apnea. Jose E. Barrera. Otolaryngl Clin Am 49-2016.

Surgical goal: Achieve the maximal advancement of the maxillo-mandibular complex and therefore improvement of the apnoea-hypopnoea index (AHI), while preserving pleasant postoperative facial aesthetics [181].

Treatment outcome: MMA increased airway dimensions by increasing the distance from the occipital base to the pogonion. An increase of this distance showed a significant correlation with an improvement in the AHI and a decreased pressure effort of the upper airway [182]. However, more evidence is still needed to draw definite conclusion related to the effect of single-jaw mandibular advancement osteotomies on pharyngeal airways [183]. Apnea hypopnea index decreased from an average of 45 to 6.2 events/hour after maxillomandibular advancement (MMA) [163, 184, 185] with a cure rate: 40% [184] and an average success rate of 66% [163, 183, 184, 186].

Surgical movement: According to previous studies, mean maxillary advancement is on average between 6 and 12 mm and more advancement for the mandible between 8 to 16 mm [184, 185, 187-189]. Although some authors of primary studies have reported the amount of surgical jaw movements, many still neglected this important information in their report. [183].

Long-term stability: surgical relapse occurred after MMA surgery ranging between 7% and 20% [190]. A successful surgical outcome was defined as an AHI decrease of >50% with <20 events/h. However, ageing and weight gain might counterbalance the positive effects of surgery in the long term. A trend towards significantly higher AHI, higher BMI, and lower sleep efficiency was found in the unsuccessful group in comparison to the successful group at long-term assessment. Younger age, lower BMI [190], mandibular retrognathia, lower preoperative AHI, no

evidence of a central sleep apnea [191] and a higher degree of maxillary advancement have been described as predictors of surgical success [190]. The addition of adjunctive procedures, such as uvulectomy, could increase surgical success [191].

PART 3: THE HYOID BONE AND UPPER AIRWAY FUNCTION

A. Hyoid bone

1. General function of the hyoid

The hyoid bone has an important function in the orofacial complex. It helps maintain upper airway patency, contributes to phonation, helps in chewing, swallowing and breathing [12, 192]. It preserves position of the head by the interrelation it creates between the mandible and the spine [12]. The hyoid bone also provides attachments to the muscles of the floor of the mouth, to the tongue, to the larynx , and to the epiglottis and pharynx [193]. The hyoid bone is involved in various prime human functions and has a strategic location and characteristics.

2. Structure

As already mentioned in chapter 1, the hyoid bone comprises a body with two lesser and two greater horns. The bony eminences present at the level of the hyoid bone are considered to be the consequence of muscular force application at the level of their attachment on that bone [194].

Muscular connections include mainly the supra and infrahyoid muscles, with a total of twenty muscles attached to the hyoid bone, ten on each side. *Ligament suspension* involves two ligaments; the stylohyoid and the thyro-hyoid ligaments. Muscular and ligamentous attachments provide equilibrium of the hyoid bone. Consequently, the position and the limit of movement of the hyoid bone are dictated by the musculature [195].

3. Hyoid - Swallowing and mastication

Reciprocity between the mandible, the hyoid and the tongue allows mastication and swallowing.

Forward and upward movement of the tongue and hyoid bone allows the transport of a bolus of food from the oral cavity to the oropharynx. This protraction takes place after the contraction of two muscles: the geniohyoid and the anterior part of the digastric muscle [196]. During swallowing, the hyoid is displaced cranially and anteriorly[197] (Figure 3.1)

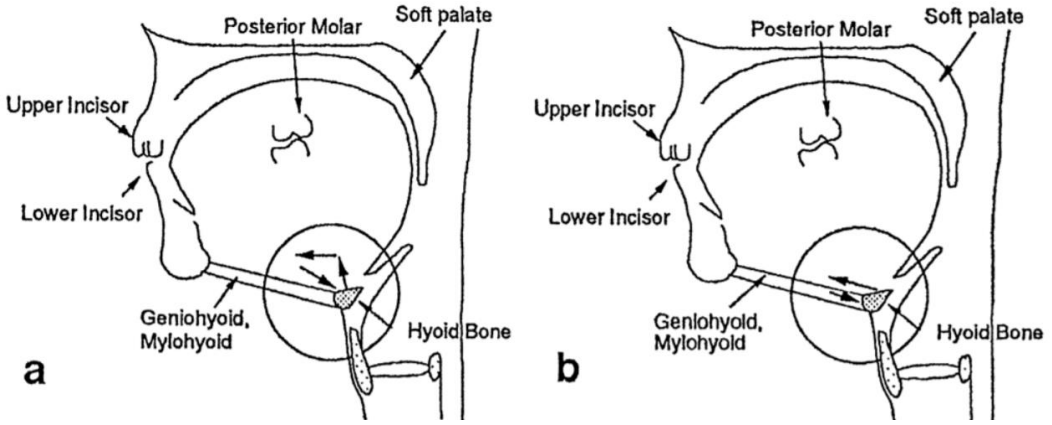


Figure 3 1: Schematic sagittal section of the oral cavity, pharynx, and larynx showing the hyoid bone and its movements during swallowing. A) Typical triangular pattern of upward displacement, then forward displacement, then return. B) Linear pattern of displacement with minimal elevation [197].

A number of studies have emphasized the importance of the hyoid movement as a component in the swallowing mechanism, with its role in the control of tongue movement [196], opening of the upper oesophageal sphincter [198], and tilting of the epiglottis [199]. Therefore, the hyoid bone movement is an important factor contributing to proper mastication and swallowing.

4. Hyoid - airway and breathing

Soft tissue volume and bony structure surrounding the upper airway can determine its dimensions.

[200]. Various studies have evaluated the correlation between the position of the hyoid bone and further craniofacial structures due to the importance of hyoid bone as a determinant of upper airway patency [201, 202].

The hyoid is linked to the epiglottis and to the posterior part of the tongue along the hyoepiglottic ligament and hyoglossus muscle respectively. This leads to a transmission of a downward force from the diaphragm to the hyoid bone during inspiration. This transfer of force occurs through the lungs, trachea, larynx, and the thyrohyoid muscles and ligament. In fact, during large breaths in humans, the hyoid bone moves anteriorly [203]. Hyoid positioning and repositioning affects the airways differently. This will be discussed in the following section.

B. Hyoid repositioning surgeries

Several hyoid bone repositioning surgeries were performed to treat OSA individuals [204, 205]. The outcome of displacement of the hyoid on upper airway flow resistance using anesthetized dogs was examined previously. Results showed that forces controlling the position of the hyoid arch can be important determinants of upper airway resistance. Accordingly, hyoid suspension was developed as a surgical procedure designed so that the hyoid bone is suspended to the inferior border of the anterior mandible. This technique was later modified by Riley et al. where he attached the hyoid anteroinferiorly to the thyroid lamina rather than suspending it from the anterior mandible. The end results of these surgeries showed significant improvement in 15

patients with OSA [206]. Anterior traction of the hyoid bone can prevent upper airway collapse when negative pressure is present [207].

1. Hyoid movement and muscular activity

Airway patency is also affected by muscular activity of the hyoid bone, not only by its position. Decrease of genioglossus tone may lead to upper airway obstruction during sleep [92] . In rabbits, geniohyoid and sternohyoid show phasic inspiratory activity that reacts to prevent upper airway collapse when negative pressure is pertained. The rabbit model was used to test the efficacy of the sternohyoid and sternothyroid muscles (upper airway muscles that connect to the hyoid bone) in maintaining the upper airway stability and patency. The effect of tension on those muscles with the animal's head at 90" is to pull the hyoid bone caudally. This caudal movement of the hyoid was associated with improved airway stability [29]. In fact, improvement in upper airway stability and dilation in the pharyngeal region seen when stimulating the genioglossus and geniohyoid muscles, was related to their effects on the hyoid bone. These muscles have the effect of shifting the hyoid bone toward the nose i.e more cranially [207]. OSA patients experience a reduction in upper airway patency in supine position. This can be due to the inability of preserving a ventral position of the hyoid bone. Moreover, this can be caused by poor coordination of hyoid muscles and the genioglossus in some patients [208]. In addition, the hyoid bone plays a pivotal role in redistributing the loads between the caudal and cranial segments and this affects the resultant upper airway geometry outcomes[86].

To summarize, the hyoid bone and its musculature play a vital role in maintaining airway patency. However, outcome of hyoid suspension surgeries in combination with other therapeutic interventions is still limited with variable success rate.

C. Importance of the hyoid bone in OSA

Patients with OSA have an inferiorly positioned hyoid bone relative to healthy individuals (Figure 3.2). This anatomical feature is consistently observed in affected individuals. This characteristic is associated with a more collapsible upper airway [3, 201, 209-211]. In healthy subjects, the hyoid is located at ~C4 [212, 213] . However, in OSA patients hyoid bone was inferiorly placed between C4 and C6. The reason could be an increased mandibular plane angle or thickened fat pads in the submental, submandibular, and pharyngeal areas [212, 213]. Moreover, the modified position of the hyoid bone leads to alteration of muscle contractile properties and force vector. The effect of a muscle when placed in a certain direction is different than when the same muscle has another orientation [214]. When the hyoid bone is positioned inferiorly, the dilatory muscles have a modified direction. This can compromise the dilation ability of these upper airway muscles [215, 216] . However, OSA patients have an increased muscular activity relative to controls when asleep but these muscles are less effective in dilating the airways [215, 216].This could be a major etiological factor for the arousal of OSA.

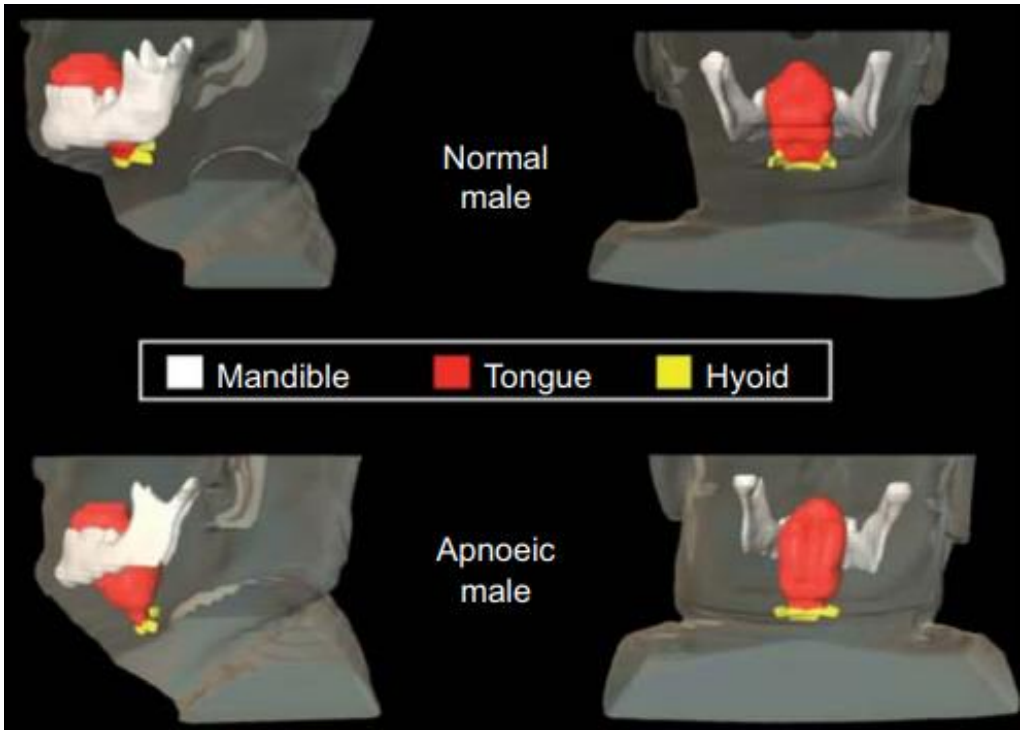


Figure 3 2: Schematic drawing, based on CT scan imaging, showing the difference between the hyoid bone position between heathy and apnoeic patient. It is placed more inferiorly and posteriorly in OSA patient. Note that the tongue volume is greater in the apnoeic patient. Adapted from Chi et al, 2011 [3].

D. Factors that alter hyoid position

Different factors affecting the upper airway dimensions and properties are often directly or indirectly related to the hyoid bone (where it becomes an intermediate transmitting forces between the supra and infra hyoid regions). Those factors include head and neck positions, tracheal displacement, obesity and mandibular advancement.

1. Head, neck and body position

Head posture is maintained due to hyoid muscle group [217]. The position of the hyoid bone in the antero-posterior plane is in correspondence with the motion of the head, jaw, and vertebrae. The position of the hyoid bone in relation to the vertebral

column had less variability than the hyoid position in relation to the maxilla and the mandible [218].

2. Tracheal displacement

Caudal tracheal displacement affects upper airways by displacing the hyoid bone caudally [88]. As a result, mechanical stability of upper airway will improve [86]. A study conducted by Amatoury et al. 2014[1] on a rabbit model showed the hyoid bone is displaced caudally from 0 to -2mm with a tracheal displacement from 0 to 10mm. Consequently, suprahyoid muscles are pulled, and peripharyngeal tissues are stretched and stiffened. This will enlarge the airway anteriorly and laterally [86]. Therefore, a change in the hyoid bone position leads to upper airway lumen geometry changes.

3. Obesity

In obese patients, fatty tissue that is deposited at the level of the neck can displace the hyoid more caudally due to tissue compression. [88].

4. Mandibular advancement

The mechanisms leading to upper airway changes with mandibular advancement, involve various interactions within the peripharyngeal tissues, including those

associated with the movement of the hyoid bone. This will be discussed in details in the following sections.

E. Mandibular advancement alone

In anesthetized and awake individuals, mandibular advancement increases the pharyngeal patency by reducing airway resistance. Closing pressure (Pclose) of upper airway is decreased in OSA patients treated with mandibular advancement. Consequently, upper airways become less collapsible [219]. Amatoury et al. investigated the effect of mandibular advancement on the upper airway collapsibility, dimensions and peri-pharyngeal tissue deformation. This was done using a 2D finite element model (FEM) of the passive rabbit upper airway and surrounding peri-pharyngeal tissues [220]. They found that incremental mandibular advancement reduces upper airway length and enlarges upper airway cross sectional area and tissue stress [221]. Amount of mandibular advancement: The specific amount of mandibular advancement that is needed to achieve optimal therapeutic outcome is still unknown. Patient selection for a positive outcome remains a challenge. The exact amount of mandibular advancement for a specific patient was never determined and many start treatment at 50–60% of maximal protrusion empirically [222]. Exaggerated mandibular advancement at 70 % increases the risk of root and bone resorption. Highest pressure is exerted on mandibular second molar. The study conducted by Lee et al, using finite element model showed that stress on the teeth and facial bones was the lowest at 40 % of maximum mandibular advancement [223]. A feedback-controlled mandibular positioner (FCMP) can be helpful in determining the exact amount of mandibular advancement that is needed according to each OSA patient. This positioner is able to

detect respiratory events and protrude the mandible according to prearranged algorithms. This has a principal clinical significance, reducing long treatment time associated with self-titration. Moreover, this test has the capability to predict not only the efficacious mandibular position, but also the outcome of oral appliance therapy. Another test that is able to do these predictions is the remotely controlled mandibular positioner (RCMP) in the polysomnographic setting [224]. It can be used to enable accurate mandibular titration and positioning during sleep [224-228]. The aim of the titration is to gradually protrude the mandible until obstructive apneas and hypopneas were terminated. The device consists of a controller that receives commands from the device software installed on the PSG computer and, in turn, activates a mandibular positioner attached to dental trays in the patient's mouth (Figure 3.3).

Figure 3 3: Scheme of the remotely controlled mandibular positioner (RCMP) that enables accurate mandibular titration and positioning during sleep [224]

The use of RCMP system in sleep centres is promising [224] but may be limited due the high cost. Another way to decrease excessive mandibular advancement and prevent potential side effects is to combine mandibular advancement with other therapeutic approaches such as hyoid repositioning interventions. This will be discussed in details in the following section.

Actually, the positive effect of mandibular advancement on upper airway can be due to the genioglossus muscle activity or to a modification in the position and motion of the hyoid bone. Mandibular advancement optimizes the genioglossus muscle activity, decreases the extraluminal tissue pressure, displaces the tongue forward and enlarges the lateral pharyngeal walls [47]. The latter was shown using dynamic magnetic resonance imaging (MRI) where upper airway tissue movement and deformation with mandibular advancement were studied in healthy participants and those with OSA[229].

F. Mandibular advancement and hyoid bone

Mandibular advancement improves airway collapsibility during sleep in a dose-dependent manner [161]. This aligns with prior studies advocating that MAS therapy primarily targets pharyngeal anatomy to improve OSA [230, 231]. After the use of MAS, sagittal linear modifications involve a 3.86% mandibular protrusion and a 1.81% anterior movement of the hyoid bone relative to the cervical vertebrae. There is also a 1.07% anterior movement of the hyoid bone relative to the maxilla. Angular movements comprise the rotation of the hyoid bone antero-superiorly by an average of -10.51%. The degree of the hyoid bone rotation may be larger than the amount of mandibular

rotation. Evaluating specific tendencies with the use of MAS will help anticipating the effects of skeletal changes to decide the best candidates for this type of treatment [232].

The hyoid bone position and motion plays a pivotal role. Amatoury et al. found that the hyoid bone moved in a cranial- anterior direction when advancing the rabbit's mandible. The hyoid bone was displaced in a cranial-anterior direction by 0.42 mm for each mm of mandibular advancement (Figure 3.4) [48]. This is in accordance with the results of other studies on human subjects [98, 107, 159, 233]. This movement will optimize the patency and stability of the upper airway as a consequence to muscles and tissue stretching. The more the hyoid bone is displaced with mandibular advancement, the better is the effect on the upper airway lumen size in awake OSA subjects. The mandible got distant from the posterior pharyngeal wall by 6.9 mm which is equivalent to 9% after a mean of 5.3mm of mandibular advancement. Moreover, the hyoid became close to the gonion by 6.9 mm and to the mandibular plane by 4.3 mm. Consequently, a 1.3mm cranial and a 1.1mm ventral repositioning was observed. The amount of change in airway dimensions bettered the mandibular advancement. The distances behind the soft palate and tongue improved by 1.0 and 0.8 mm, respectively [107]. In fact, the suprahyoid muscles that link the hyoid to the mandible are likely stretched when the mandible is repositioned forward. The hyoid and the thyroid cartilage are then displaced cranially and tissues are stiffened in that area [48]. Thus the hyoid bone plays a crucial role in delivering the mandibular advancement effect to the caudal peripharyngeal tissues. Moreover, the movement and mobility of the hyoid bone is to be considered. Impeding hyoid bone movement limits the load transfer from upper to lower tissues that should occur with mandibular advancement [163, 234] . This induces the stiffening of upper airways. Until today, it is still not well established whether movement of the

hyoid bone more caudally or cranially with mandibular advancement contributes to improvements with MAS therapy [163, 234] .

Moreover, the baseline position of the hyoid bone, which determines the effectiveness of the upper airway dilator muscles, might also affect the mandibular advancement outcomes. Thus, the hyoid bone original position might be a potential factor for mandibular advancement treatment success in OSA patients. After mandibular advancement, patients can have significant ventral-cranial elevation of the hyoid bone by 8.8±1.8mm (from 25.3 ± 7.8 mm without MAS to 16.5 ± 9.6 mm with MAS), thus acquiring a more normal hyoid position [235]. The distance between the hyoid bone and the mandible (MP-H) decreased significantly by 9.0±0.1 for patients treated with MAS [236]. Significant decrease in AHI during sleep were obtained by establishing a new anterosuperior position of the hyoid bone relative to the mandibular plane. It is postulated that either the reduction in the perpendicular distance between the hyoid bone and the mandibular plane or the maintenance of an adequate hyoid position while asleep is the mechanism whereby upper airway patency is achieved[235].The precise influence of hyoid repositioning in concomitance with mandibular relocation on upper airway function is still not very well defined.

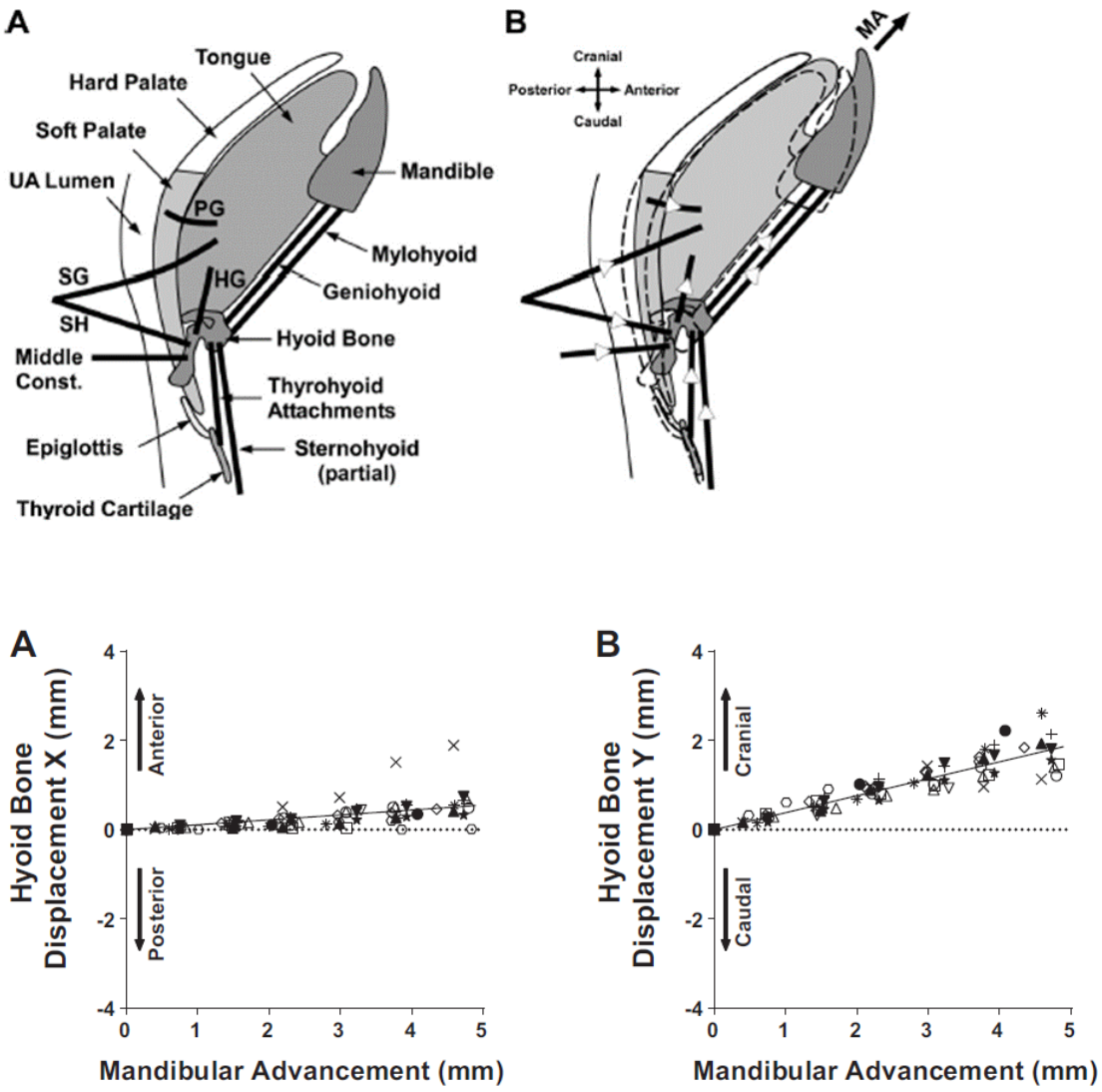


Figure 3 4: Scheme and graphs showing the effect of mandibular advancement on the displacement of the hyoid bone in a passive rabbit model. The hyoid is displaced anteriorly and cranially with the progressive amounts of mandibular advancement. Adapted from Amatoury et al. 2014 [48] .

In addition, the influence of mandibular advancement on hyoid position is debatable. As previously mentioned and according to earlier studies in

humans (Battagel *et al*, 1999, Liu *et al*, 2000, Almeida *et al*, 2006, Poon *et al*., 2008 and Doff *et al*, 2009) the hyoid bone is repositioned cranially after mandibular advancement. However, this is controversial with the results of the study conducted by Medina et al 2020. Their aim was to assess the position of the hyoid bone in Cl II malocclusion patients that are treated using a mandibular advancement splint (the activator). They found that the hyoid bone is displaced ventrally but also caudally after mandibular advancement [237]. The reason behind this variation is unknown and further investigations are needed to determine the exact movement of the hyoid bone as a consequence to mandibular advancement therapy.

Combined therapy: Repositioning both the mandible and the hyoid bone together can lead to better therapeutic outcome. This is possible due to combined therapy. In fact, OSA is a heterogenous disorder leading to the necessity of a tailored therapy that is adapted to the pathophysiology of each patient. Combined therapy is needed to enhance therapeutic outcome and to tackle efficacy and compliance problems [238]. For instance, a combination of MAS and CPAP use is a viable option to decrease PAP requirement [239-241]. A study conducted by Cistulli et al 2020 [242] examined the dose-dependent effects of mandibular advancement on optimal CPAP requirements in OSA patients. A research polysomnography(PSG) was performed to determine optimal CPAP requirements at five predetermined mandibular advancement positions (0% 'habitual bite' and 25, 50, 75 and 100% of the maximal mandibular advancement). A second PSG [243] was performed approximately 6 weeks after initiation of MAS therapy to determine treatment outcome. The average reduction in optimal CPAP requirements from 0 to 100% advancement was 4.0 ± 2.7 cmH_2O. This highlights the positive outcome of incremental mandibular advancement and the importance of

combined therapy. A physiologic study showed that velopharyngeal resistance was reduced in the mandibular advancement/nasal mask combination as compared to CPAP alone [244]. Moreover, MAS can be used in concomitance with positional therapy to have a better result [245]. Surgical treatments can be merged with the use of MAS such as uvulopalatopharyngoplasty (UPPP) and tracheal displacement [246]. In fact, greater long-term control of OSA was found with the combination approach compared to surgery alone [247]. Other surgical approaches can be combined with MAS such as hyoid bone repositioning; however, this option has not been thoroughly assessed and has received little attention in the literature [248]. This can be beneficial, that's why more studies on combined therapeutic approaches are needed [249].

CHAPTER II

HYPOTHESIS AND AIMS

A. Summary and rationale

The upper airway is a complex structure [95] that includes both dynamic (muscles, neuromuscular control) and static (bone, soft tissues) components. Equilibrium between collapsing and dilating forces helps in maintaining airway patency. Sleep breathing disorders such as obstructive sleep apnea (OSA) are the consequence of loss of equilibrium making the upper airway prone to collapse. In this study, we examine the combined influence of mandibular advancement and hyoid bone position on UA collapsibility. Final outcomes help understand the role of the hyoid bone repositioning and mandibular advancement therapy on upper airway collapsibility, and suggest potential combined therapy approaches to treat this serious condition.

B. **Aims**

The main aim of this thesis is to understand how the position of the mandibular and hyoid bone influence upper airway patency from a quantitative perspective. Therefore, invasive interventions are needed that cannot be achieved in humans. The rabbit has a similar upper airway structure to humans especially the fact that they have a freely suspended hyoid bone (unlike most non-primates where the hyoid bone is fixed to the surrounding structures). The model has been used in multiple studies in respiratory physiology and was found to be ideal with outcomes applicable to humans. In addition, using an animal model reduces the number of subjects needed because of the decreased inter-individual variations in animals.

The specific aims of this project are to determine:

1. The effect of mandibular advancement on upper airway patency
2. The effect of hyoid repositioning combined with mandibular advancement on upper airway patency

We quantify the upper airway collapsibility using the closing pressure metric, Pclose, with different mandibular and hyoid bone positions following different angles covering the caudal, cranial, anterior, and 45degrees caudally and cranially with 2 and 4 millimetres of mandibular advancement.

CHAPTER III

MATERIAL AND METHODS

A. Subjects

Studies were performed on a total of 12 adult, male, New Zealand White rabbits with the following characteristics: weight= 2.88 Kg ± 0.18, neck circumference: 15.19 cm ± 2.51 and height: 38 cm ± 2.36. Rabbits were bred and housed in the animal care facility at the American University of Beirut.

B. Ethics

The protocol was approved by the American University of Beirut Institutional Animal Care and Use Committee.

C. Anaesthesia

Anaesthesia was initially induced with an intramuscular injection of ketamine (35mg/kg) and xylazine (5mg/kg) and then maintained with a continuous intravenous infusion (through an ear vein) of ketamine (15 mg/kg/hr) and xylazine (4.5 mg/kg/hr) at an infusion rate of 0.025 ml/min/kg (Figure III.1). The rabbit's ear was cannulated for anaesthetic maintenance using 24 G cannula in the lateral auricular vein no later than 30 minutes after the last intramuscular dose. The other ear was also cannulated as a spare. Animals were euthanized at the completion of each study by an overdose of the intravenous ketamine/xylazine anaesthetic.

The details for the anthropometric parameters and anaesthetic dose for each rabbit are shown in table III 1.

Table III 1. Anthropometric parameters of the rabbits and anesthetic details

D. Preparation and surgery

#	Name	Experiment	Weight (kg)	Neck circumference	Height	Drug dose ketamine/xylazine	Infusion rate
1	JA01	JA01_HYMA01	3.10	16.5	37	1.3ml/0.75ml	5 ml/hr
2	JA02	JA02_HYMA02	3	16	36	1.1ml/0.75ml	4.5ml/hr
3	JA03	JA03_HYMA03	2.9	15.9	36.5	1.1ml/0.75ml	4.5ml/hr
4	JA04	JA04_HYMA04	3	16.3	36.5	1.1ml/0.75ml	4.5 ml/hr
5	JA05	JA05_HYMA05	2.80	15.8	41	1.1ml/0.75ml	4.2 ml/hr
6	JA06	JA06_HYMA06	3.1	15	34	1.3ml/0.75ml	4.65 ml/hr
7	JA07	JA07_HYMA07	2.80	15.5	42	1.1ml/0.75 ml	5 ml/hr
8	JA08	JA08_HYMA08	2.60	16.8	40	1ml/0.75ml	4ml/hr
9	JA09	JA09_HYMA09	2.80	15	37	1.11ml/0.75ml	4.5ml/hr
10	JA10	JA10_HYMA10	2.80	7.5	37	1.11ml/0.75ml	4.5ml/hr
11	JA11	JA11_HYMA11	2.60	15	40	1ml/0.75ml	4.5ml/hr
12	JA12	JA12_HYMA12	3.10	17	39	1.3ml/0.75ml	5 ml/hr

The rabbit was fasted for 12 hours prior to surgery. Before the start of every surgery, the anaesthetic infusion, catheter instrumentation and saline infusion set up were

prepared and the pressure transducers were checked for proper calibration. The animal was then transported from the animal housing facility to the SRB building, JA laboratory where the initial anaesthetic intramuscular injection was done.

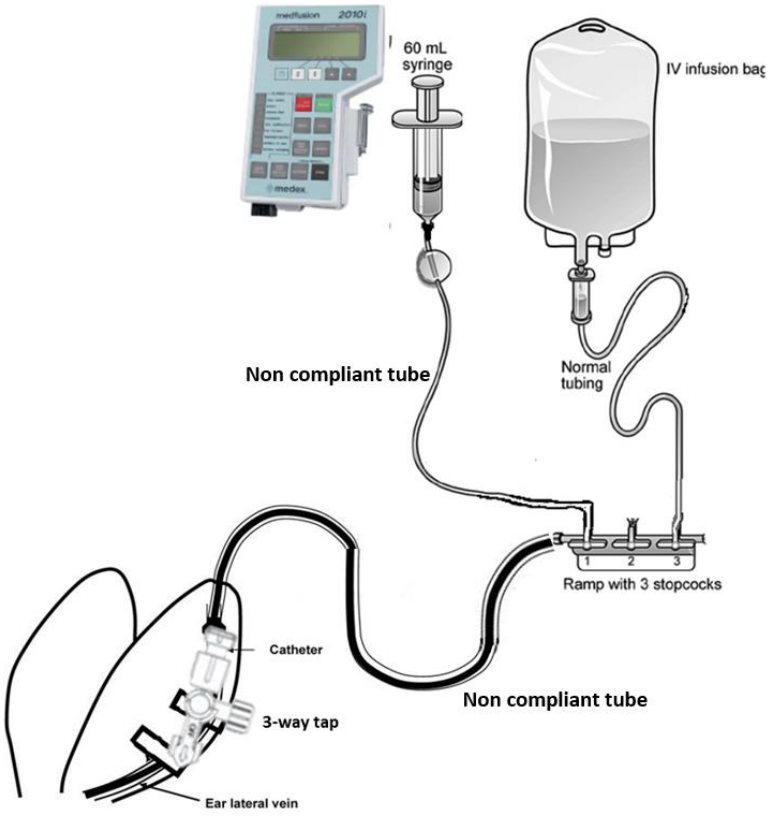


Figure III 1: Schematic drawing showing the anaesthetic and saline infusion set up, prepared the morning of the surgery. The saline bag and the anaesthetic syringe are connected to a ramp with 3 stopcocks from which a non-compliant tube emerges and is inserted in the 3-way tap which forms the extension of the catheter.

E. Experimental Setup

The rabbits were supine positioned on a surgical platform. The upper limbs were secured with tape to the bench in such a way as not to stress the thorax and avoid injury

to the resting heart rate and breathing. The head/neck position was controlled through an adjustable head rest and measured using a protractor to have a line drawn from the tragus to the external nares at 50 degrees to the horizontal.

A skin incision was made on the ventral surface of the neck and blunt dissection was used to expose the trachea. The baseline position of the trachea, taken between the fourth and fifth tracheal cartilage rings, was marked on the fixed experimental platform at the end of expiration. The skin incision extended to the level of the symphysis at the lower border of the mandible in order to expose the hyoid bone for the setup of the hyoid bone re-positioning device.

The upper airway was isolated via complete surgical transection of the trachea (between the 3rd and 4^{th} tracheal cartilage rings). Thus, no airflow was detected though the upper airway (rabbits breathing via caudal trachea) using diathermy (Valleylab Force 2 Electrosurgical Unit) (Figure III.2). An L- shaped tube was inserted and secured (2.0 prolene, Johnson & Johnson, Livingston, UK) into the caudal tracheal segment, through which the rabbit breathed spontaneously. After transecting the trachea, the cranial segment was brought back to its baseline position by placing a suture around the cranial L shaped tube and connecting it to a vertical stand at the end of the bench to control the tracheal position.

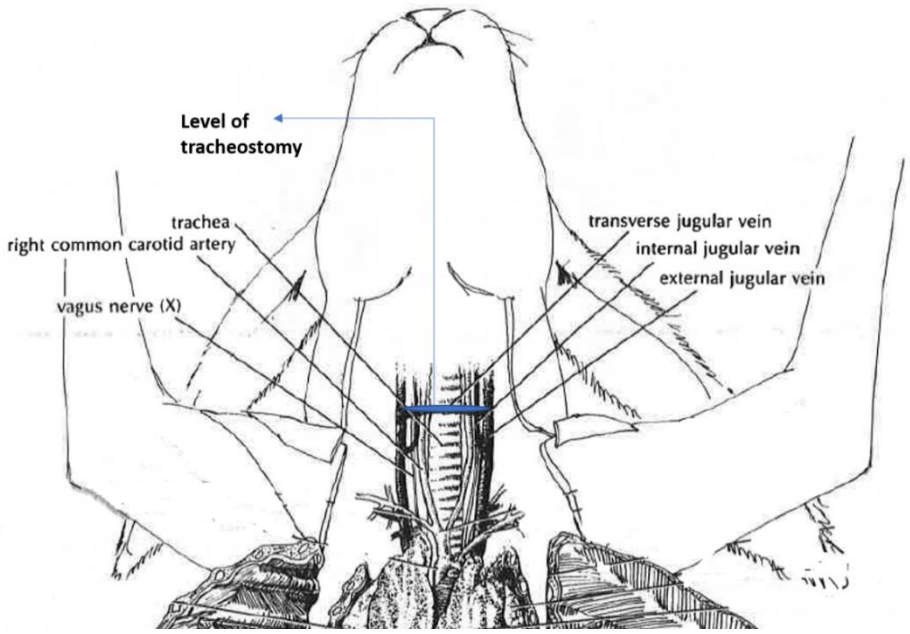


Figure III 2: Schematic drawing of the rabbit's trachea. The tracheostomy is performed between the 3rd and the 4th tracheal cartilaginous rings. Adapted and modified from Wingerd, 1985 [108].

The L – shaped tube was used to connect to the flow and pressure measuring devices. A custom-made L shaped tube was inserted into the cranial tracheal segment and secured using a suture (2.0 prolene, Johnson & Johnson, Livingston, UK). This tube was connected to a calibrated syringe and 100 cm volume extension (1*2.5mm), and a pressure transducer to measure upper airway pressure (Pua).

A small modified conical animal anaesthetic mask (GaleMed VM-2, GaleMed, Taiwan) was fitted to the rabbit's snout to achieve a closed upper airway system for application of upper airway intra-luminal pressure and measurement of mask pressure. To achieve a proper seal around the rabbit's snout, an inflatable sleeve was incorporated into the mask (Figure III.3). The inner sleeve consisted of a rubber tube segment (Penrose, flat size = 25mm).

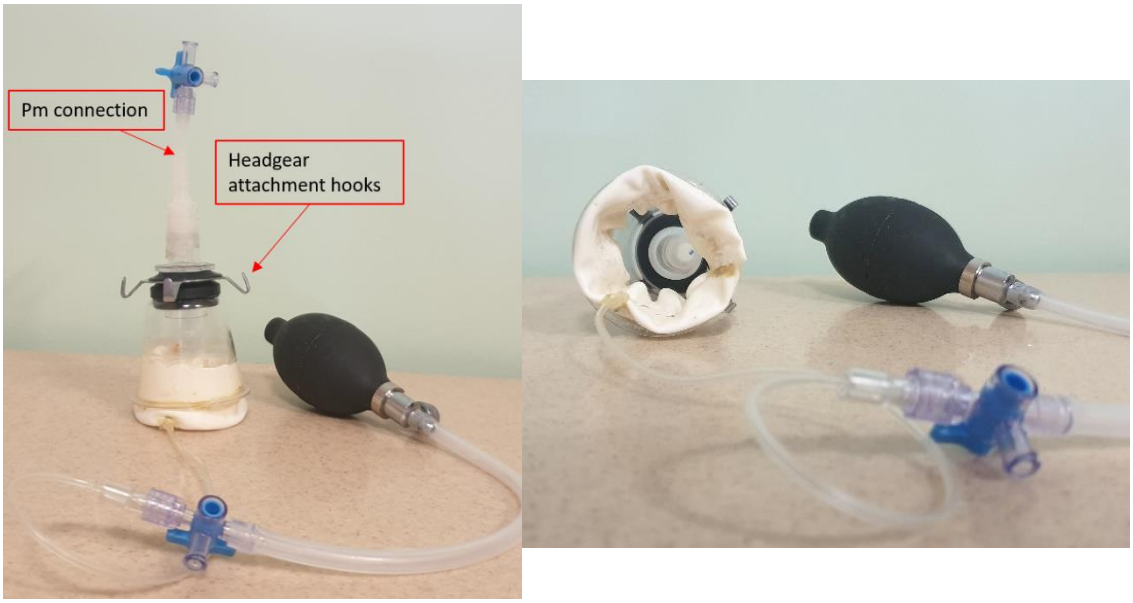


Figure III 3: A- the mask is shown in the vertical position with the connection tubing. B- the mask is shown from the bottom view with the Penrose tube deflated. The sleeve was inflated using a sphygmomanometer pump to provide seal when fitted on the rabbit's snout. Hooks around the top of the mask were used to attach a rubber custom-made headgear. A connection for the measurement of mask pressure is also available.

A pressure transducer (mask pressure; Pmask) was connected to the mask to measure the pressure at the level of the rabbit's face. A complete mask seal was ensured by pressurizing the system through a syringe and noting any pressure leak (by increasing the pressure in the upper airway (Pua), it was detected similarly at the level of the mask and maintained over 20 seconds; Pmask = Pua) (Figure III.4).

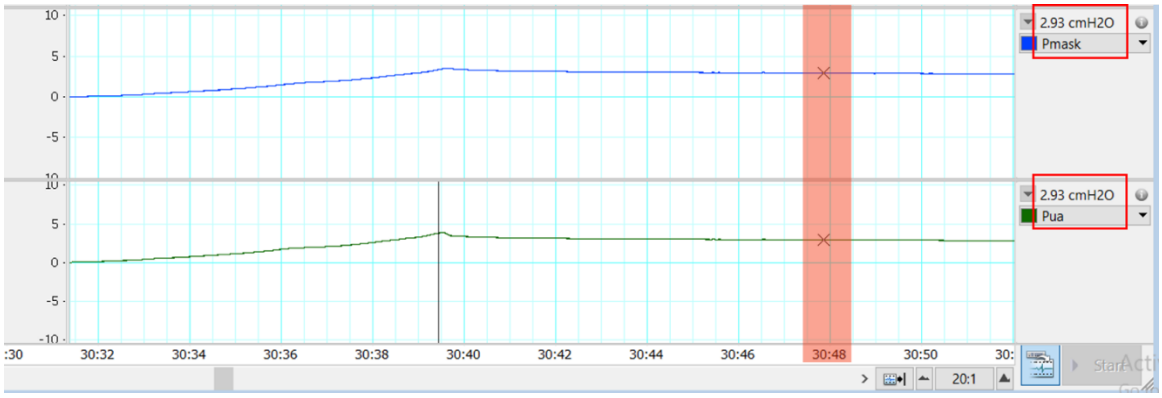


Figure III 4: Recordings of Pua and Pmask while introducing positive pressure using a calibrated syringe in the upper airway. The pressure is the same and it is maintained (Pua = Pmask = 2.93 cmH_2O) as highlighted in red on the graph therefore no leakage in the upper airway system was detected.

F. Monitoring

Basic vital signals were monitored throughout the experiment to ensure that the rabbit was in stable anesthetized state, and later to help confirm euthanasia.

- Electrocardiogram: A 3-lead electrocardiogram (ECG) was used throughout the study to monitor cardiac rhythm and heart rate. ECG electrodes were attached to the rabbit's paws after scrubbing them using sandpaper and placing ECG gel to reduce impendence [upper left paw (red -), lower left (black +), lower right (ground)].

- Tracheal pressure and flow: Tracheal pressure was measured from the caudal tracheal segment using a pressure transducer (Validyne DP45–32; Validyne Engineering, Northridge, CA) connected to the trachea via an L-shaped cannula, 3-way tap and extension tubing (Microbore hard extension set, length = 600mm, internal diameter= 1mm, outer diameter= 2.5mm). The pressure transducer was coupled to a control unit (Validyne CD72-4) (Figure III.5).

-An oximeter (ML325/AC) was used to measure the oxygen saturation level during the surgical intervention.

Figure III 5: Baseline recording of acquired signals before the start of the protocol. Electrocardiography (ECG), Pressure at the level of the mask (Pmask), Pressure at the level of the upper airway (Pua), tracheal pressure (Ptrach), Heart rate (HR).

G. Data acquisition

The mandible was advanced anteriorly from 0 to 4mm at 2mm increments. Moreover, the hyoid bone was displaced from 0 to 4mm at 2mm increments (anteriorly, cranially, caudally, antero-cranially and caudally) at every mandibular repositioning increment. At each position, the upper airway pressure was reduced progressively by pulling the syringe, creating a negative pressure inside the airway system. Since it is a closed system, the pressure detected at the level of the mask (Pm) is the same as the pressure induced at the level of the trachea (Pua). Pua and Pm were monitored carefully

until reaching the point pressure leading to upper airway closure. This point is when the pressures upstream and downstream diverge, where Pua continues to decease though Pmask no longer changes (Pm ǂ Pua). The minimal pressure value reached by Pm before diverging from Pua is equal to the closing pressure of the upper airway (Pclose). (Figures III.6 and III.7).

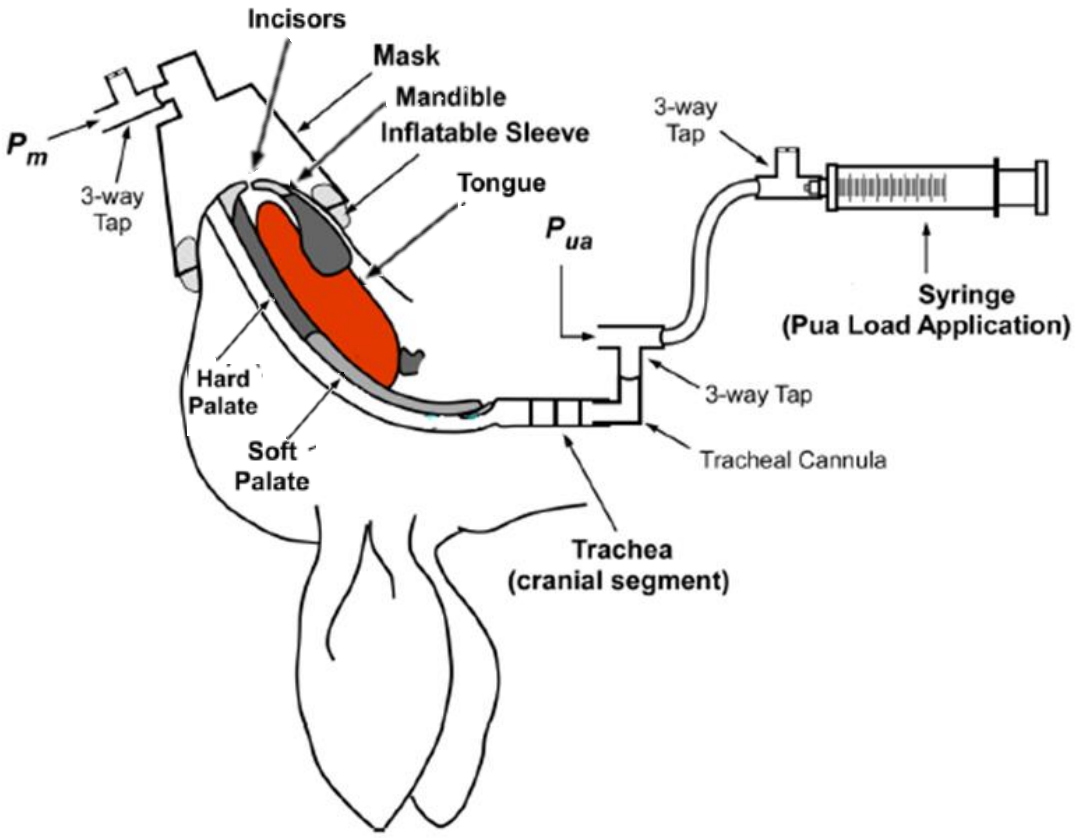


Figure III 6: Schematic of experimental setup for upper airway intra-luminal pressure application. The syringe is pulled to create a negative pressure in the upper airway, which is detected at the level of the mask. Adapted from J. Amatoury, PHd thesis, 2012.

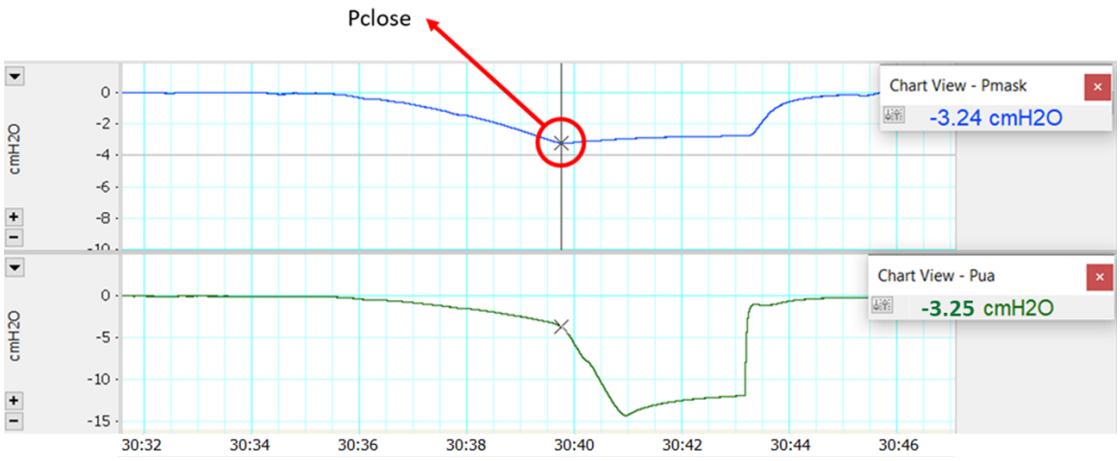


Figure III 7: Screenshot of the Pmask and Pua recordings showing the Pclose value obtained when the hyoid was still at baseline position. Before the closure of the upper airway, Pmask and Pua decreased at the same rate as the negative pressure was applied in the upper airway. Notice the sudden drop in Pua once the upper airway was closed while Pmask was stabilized at -3.24 cmH2O until the upper airway was reopened to atmospheric pressure again, then Pmask and Pua were brought back to 0 cmH_2O (the 3-way tap connected to the calibrated syringe is opened to the atmosphere).

A starting point measurement was obtained by applying a negative pressure in the upper airway until reaching the closing pressure (Pmask differing from the Pua). This application was repeated 3 times before setting up the hyoid and mandibular devices.

H. Devices set-up

1. Hyoid device set-up

1. An orthodontic miniscrew (RMO® Dual-Top, 2mm x 8mm) was inserted using a screwdriver into the body of the hyoid bone. The greater horns of the hyoid bone were held with surgical hemostats and rotated anteriorly to facilitate the placement of the miniscew at the center of the hyoid body. The

miniscrew was placed horizontally, at the midline of the hyoid body at the level of the basihyoideum.

The hyoid device consisted of a horizontal beam supported by 4 vertical stands. A clamp extending from the horizontal part was attached to the hyoid through the miniscrew and stability was tested to detect any movement between the clamp, the miniscrew and the hyoid.

2. Two calipers, with digital display, perpendicular to each other were used to reposition the hyoid bone: (Figure III.8).

- a vertical caliper was used to control anterior-posterior position of the hyoid.
- a horizontal caliper was used to control the craniocaudal position of the hyoid.

The device was fixed to the Perspex bench via four vertical stands that were held rigidly to the surgical platform by tightening the corresponding screws around them.

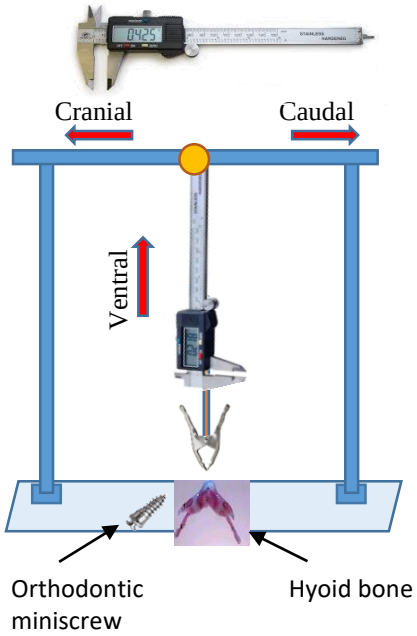


Figure III 8: Hyoid device setup

2. Mandibular device set-up

1. A mandibular advancement splint was custom made at the AUBMC dental laboratory, division of Orthodontics and Dentofacial Orthopedics. The device was fabricated on plaster models of the maxilla and the mandible of the rabbit. The impression of the 3D printed design was taken using alginate impression material and poured in white plaster. The appliance was fabricated using cold-curing orthodontic acrylic resin (Vertex-Dental, AOPP2201000, shade 22, Netherland) and an expansion orthodontic screw (Leone, A0890, Italy) that could move the mandible in small gradual increments (0.2mm), precisely and steadily, in the anterior direction (Figure III.9). The MAS attaches to the lower and upper incisors using glass ionomer luting cement (3M ESPE, self-curing, Germany). The mandibular advancement splint was fitted such that the angle of mandibular advancement was at 70° to the horizontal. Turning the screw in a clockwise direction displaced the mandible forward. Each full turn of the screw achieved 0.2mm of advancement.
2. A minimal opening of the bite followed the insertion of the splint. Bite opening was measured using two fixed and stable landmarks and a caliper. Two fixed landmarks were marked on the maxillary and mandibular jaws/teeth and bite opening was measured before and after insertion of the splint using the caliper in the vertical direction. This lead to the necessity of trimming from the incisal edge of the upper and lower incisors to decrease bite opening and to ensure a better fitting of the incisors inside the device. Trimming of the incisors was done using a

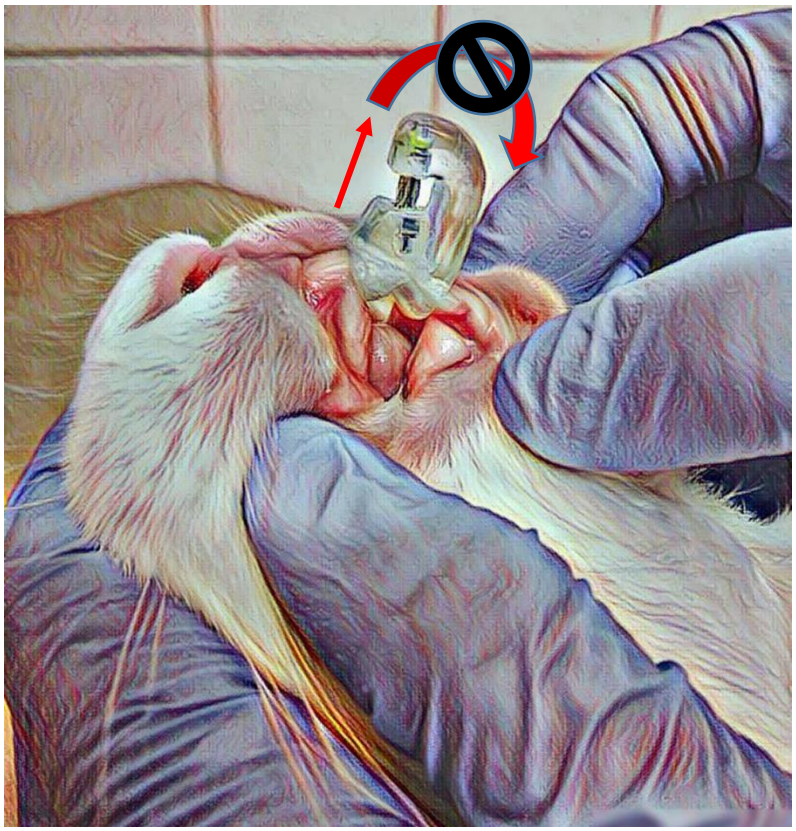
rotary tool and cut-off wheel (diameter x thickness=23.8x0.06mm). The amount of trimming was quantified with the caliper.

3. The screw of the mandibular advancement device allows a total of 7mm of advancement with 0.2 mm increments. Three positions were taken into consideration: baseline position, 2mm and 4mm advancement.

   A marking was made in the device to register the baseline position.

   -To advance the mandible by 2 mm, 10 screw turns were required.

   -To advance the mandible by 4 mm, 20 screw turns were required.



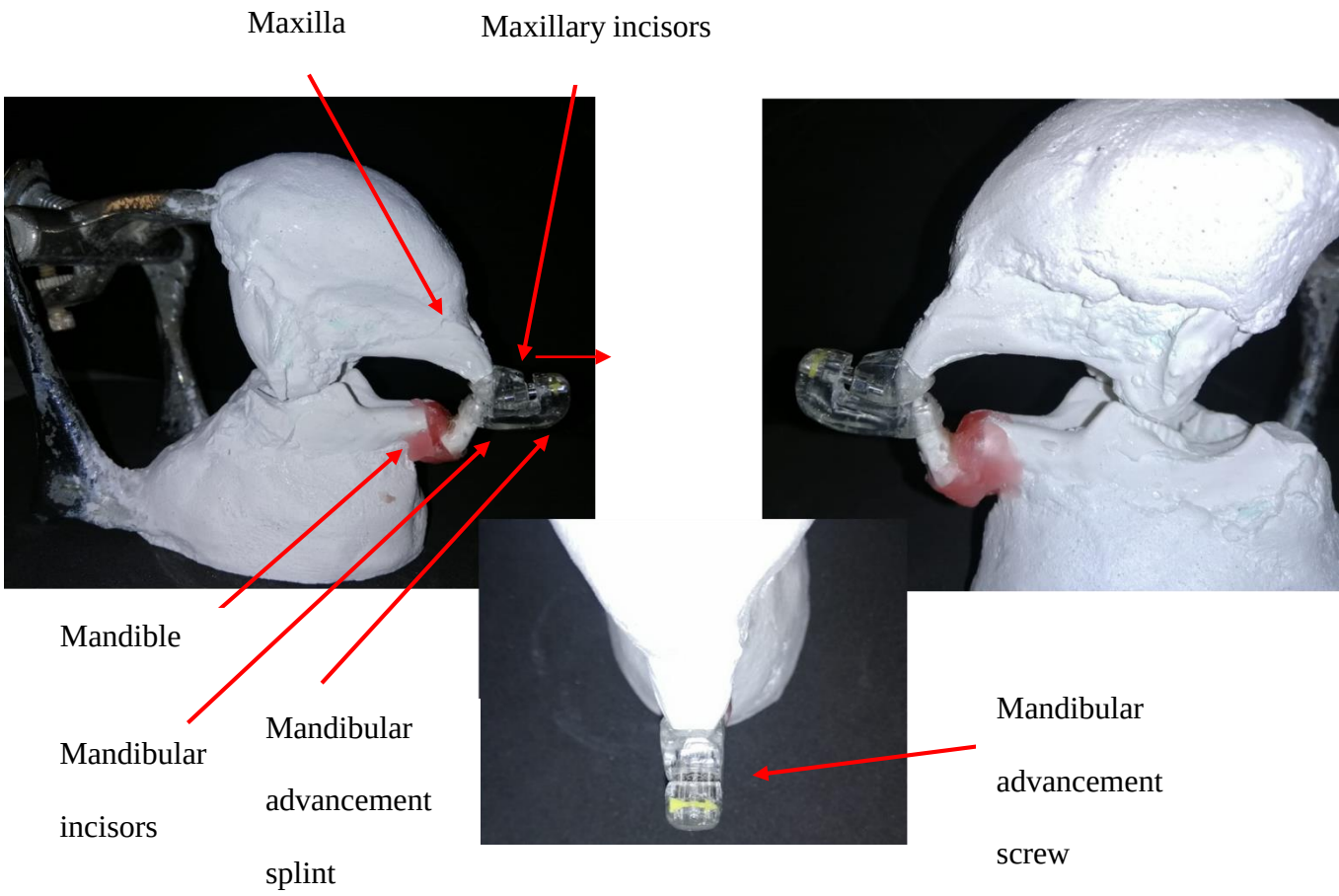


Figure III 9: Mandibular advancement splint design.

I. Protocol

Mandibular advancement and hyoid repositioning

The mandible was advanced at an angle of 70° to the horizontal in 0.2mm increments from 0 to 4 mm (0,2,4 mm). For each mandibular advancement level, the hyoid was repositioned (Figure III.10) in the following sequence:

- Anterior: 0 to 4 at 2 mm increments anteriorly (0-2-4mm)

- Cranial: 0 to 4 at 2 mm increments cranially (0-2-4mm)

- Caudal: 0 to 4 at 2 mm increments caudally (0-2-4mm)

- Ant- cranial 45°: 0 to 4 at 2 mm increments antero-cranial (2mm anteriorly with 2mm cranially then 4mm increments)

- Ant- caudal 45°: 0 to 4 at 2 mm increments antero-caudal (2mm anteriorly with 2mm caudally then 4mm increments).

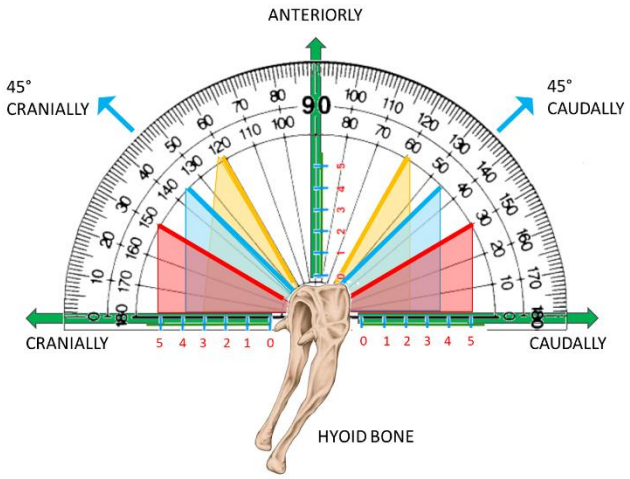


Figure III 10: Drawing showing the angles of hyoid bone movement: cranially, caudally, anteriorly, ant-caudal 45 and ant-cranial 45. The hyoid bone was displaced from 0 to 4mm at 2mm increments in the corresponding directions.

The whole cycle of measurements was repeated 3 times. For all interventions and runs, once steady state conditions were present, all physiological signals were recorded.

Upon completion

1. The rabbit was euthanized using an overdose of ketamine/xylazine
2. The euthanization was confirmed by:

   -ECG signal

   -Checking reflexes

-Clamping then releasing the tongue to determine if an arterial fill occurs

-Gel like look eyes

3. The catheters were carefully removed and cleaned as per instructions and stored in corresponding boxes
4. All devices connected to the rabbit were removed
5. The rabbit was bagged and placed in the animal house freezer.

J. Data and statistical analysis

All physiological signals were obtained using power lab and read on Lab Chart 8 (Adinstruments Ltd, Colorado, USA). The primary outcome P close measurement was averaged for each rabbit for all 3 runs. Then, group averaged data for all rabbits were represented as mean ± SD. Absolute and delta measurements were obtained. The delta value, the difference between the actual measurement and baseline, was used to track changes after mandibular and hyoid repositioning relative to baseline.

A repeated measure three-way ANOVA (SPSS v24, IBM) was used to analyse the effect of the two independent variables (hyoid directions/increments and mandibular advancement increments) in each intervention (mandibular advancement and hyoid repositioning anteriorly, cranially, caudally, ant-cranially 45° and ant-caudally 45°) on the outcome variable Pclose (dependant variable) as well as the interaction between the independent variables.

Mauchly's Test of Sphericity was used to test the assumption that the relationship between the different pairs of conditions was similar. The test of within-subjects was used to evaluate changes resulting from the independent variable. The pairwise comparison was employed to assess which pairs of conditions significantly differed

from one another. The Holm-Bonferroni post hoc was performed to assess changes in key outcomes with the hyoid and mandibular movements. Statistical significance for all the analyses was inferred for P less than 0.05.

CHAPTER IV

RESULTS

A. Interaction between MA, hyoid repositioning direction and increment

The interaction between MA, hyoid direction and increment was highly significant ($p<0.001$). Moreover, the interactions between MA and hyoid direction, MA and hyoid increment and between hyoid direction and increment were also significant ($p<0.001$).

B. Effect of MA, hyoid repositioning direction and increment on P close

The test of within-subject effects showed significant main effects of hyoid direction on the mean Pclose, of hyoid increment on mean Pclose and of MA on mean Pclose ($p<0.001$). These relations were proven using the assumption of sphericity test that stipulates that the relationship between the different pairs of conditions is similar. Also the combination of MA, hyoid repositioning increment and direction had a significant main effect on Pclose ($p=0.000$).

C. Hyoid repositioning direction and increment

A statistically significant difference ($p<0.001$) was found for the hyoid displacement direction between anterior and cranial, anterior and caudal, cranial and ant-cranial, cranial and ant-caudal ($p=0.001$), caudal and ant-cranial, caudal and ant-caudal (Table IV.1). The average absolute P close was lowered for anterior, ant-cranial and ant-caudal compared to cranial and caudal hyoid repositioning. For the same hyoid

increment, i.e. 2mm hyoid repositioning with no MA, the absolute P close obtained with anterior repositioning was of -5.3 cm H2O (Figure IV.1 A), compared to P close values of -4.1 and -4 cmH2O for cranial (Figure IV.1 D) and caudal (Figure IV.1 G) hyoid repositioning, respectively. The absolute P close obtained with ant-cranial 45° repositioning was -5.1 cmH2O (Figure IV.1 J) and -5.2 cmH2O with the ant-caudal 45° hyoid repositioning (Figure IV.1 M). No statistically significant difference was observed between cranial and caudal ($p=0.532>0.05$) or between anterior, ant-cranial and ant-caudal ($p=1.000$).

A highly statistical significant difference in Pclose ($p=0.000$) was observed for hyoid increments between 0 and 2mm, 2 and 4mm and 0 and 4mm for anterior, ant-cranial 45° and ant-caudal 45° hyoid repositioning (Table IV.2). With the increase of hyoid repositioning, Pclose decreased progressively ($p=0.000$). The Pclose obtained with 0 and 2mm hyoid repositioning was significantly higher at all incremental levels compared to 4mm of hyoid dispalcement ($p=0.000$). Pclose decreased progressively with increasing anterior, ant-cranial 45° and ant-caudal 45° hyoid bone repositioning for all rabbits ($p<0.001$). The average absolute Pclose obtained with the 4mm anterior hyoid repositioning (MA=0) was -6.6 cmH2O compared to the average absolute Pclose of -4.1 cmH2O and -5.3cm H2O when the hyoid was repositioned anteriorly by 0 and 2mm, respectively (Figure IV.1 A). The average absolute Pclose observed with ant-cranial 45° hyoid repositioning (4mm) was -6.9 cmH2O compared to the corresponding Pclose of -4 cmH2O when the hyoid was not repositioned (Figure IV.1 J). The average absolute Pclose with ant-caudal 45° hyoid repositioning (4mm) was -6.4 cmH2O compared to the average absolute Pclose of -4 cmH2O with no hyoid repositioning (Figure IV.1 M).

When the hyoid was moved cranially or caudally, Pclose remained the same, the average absolute P close obtained when the hyoid was repositioned cranially by 0,2 and 4mm was unchanged at a value of -4.1cm H2O (Figure IV.1 D). The average absolute P close observed when the hyoid was repositioned caudally by 0,2 and 4mm also was unchanged at a value of -4cm H2O (Figure IV.1 G).

The improvement (i.e. decrease) in Pclose following anterior hyoid repositioning was not significantly different from that obtained with the ant-caudal 45° and ant-cranial 45° repositioning ($p>0.05$). Furthermore, the ant-caudal 45° hyoid repositioning was not significantly different than the ant-cranial 45° repositioning ($p=1.000$). On the other hand, Pclose was significantly different when anterior, ant-caudal 45° and ant-cranial 45° hyoid advancements were compared to the cranial and caudal directions.

Table IV 1: Pairwise comparisons to determine which pairs of conditions significantly differ from one another for hyoid repositioning direction

Pairwise Comparisons

Measure: MEASURE_1

(I) Direction	(J) Direction	Mean Difference (I-J)	Std. Error	Sig.[b]	95% Confidence Interval for Difference[b]	
					Lower Bound	Upper Bound
Ant	Cran	-1.253*	.169	0.000	-1.843	-.663
	Caud	-1.381*	.173	0.000	-1.986	-.776
	Ant-cran	-.026	.108	1.000	-.403	.351
	Ant-caud	-.204	.130	1.000	-.660	.252
Cran	Ant	1.253*	.169	0.000	.663	1.843
	Caud	-.128	.059	0.532	-.336	.079
	Ant-cran	1.227*	.179	0.000	.600	1.855
	Ant-caud	1.049*	.174	0.001	.441	1.657
Caud	Ant	1.381*	.173	0.000	.776	1.986
	Cran	.128	.059	0.532	-.079	.336
	Ant-cran	1.356*	.170	0.000	.761	1.951
	Ant-caud	1.177*	.147	0.000	.662	1.693
Ant-cran	Ant	.026	.108	1.000	-.351	.403
	Cran	-1.227*	.179	0.000	-1.855	-.600
	Caud	-1.356*	.170	0.000	-1.951	-.761
	Ant-caud	-.178	.092	0.794	-.500	.144
Ant-caud	Ant	.204	.130	1.000	-.252	.660
	Cran	-1.049*	.174	0.001	-1.657	-.441
	Caud	-1.177*	.147	0.000	-1.693	-.662
	Ant-cran	.178	.092	0.794	-.144	.500

Based on estimated marginal means

*. The mean difference is significant at the .05 level.

b. Adjustment for multiple comparisons: Bonferroni.

Table IV 2: Pairwise comparisons to determine which pairs of conditions significantly differ from one another for hyoid repositioning increments

Pairwise Comparisons

Measure: MEASURE_1

(I) Disp	(J) Disp	Mean Difference (I-J)	Std. Error	Sig.[b]	95% Confidence Interval for Difference[b]	
					Lower Bound	Upper Bound
0	2	.756*	.099	0.000	.476	1.036
	4	1.490*	.187	0.000	.963	2.017
2	0	-.756*	.099	0.000	-1.036	-.476
	4	.734*	.101	0.000	.450	1.019
4	0	-1.490*	.187	0.000	-2.017	-.963
	2	-.734*	.101	0.000	-1.019	-.450

Based on estimated marginal means

*. The mean difference is significant at the .05 level.

b. Adjustment for multiple comparisons: Bonferroni.

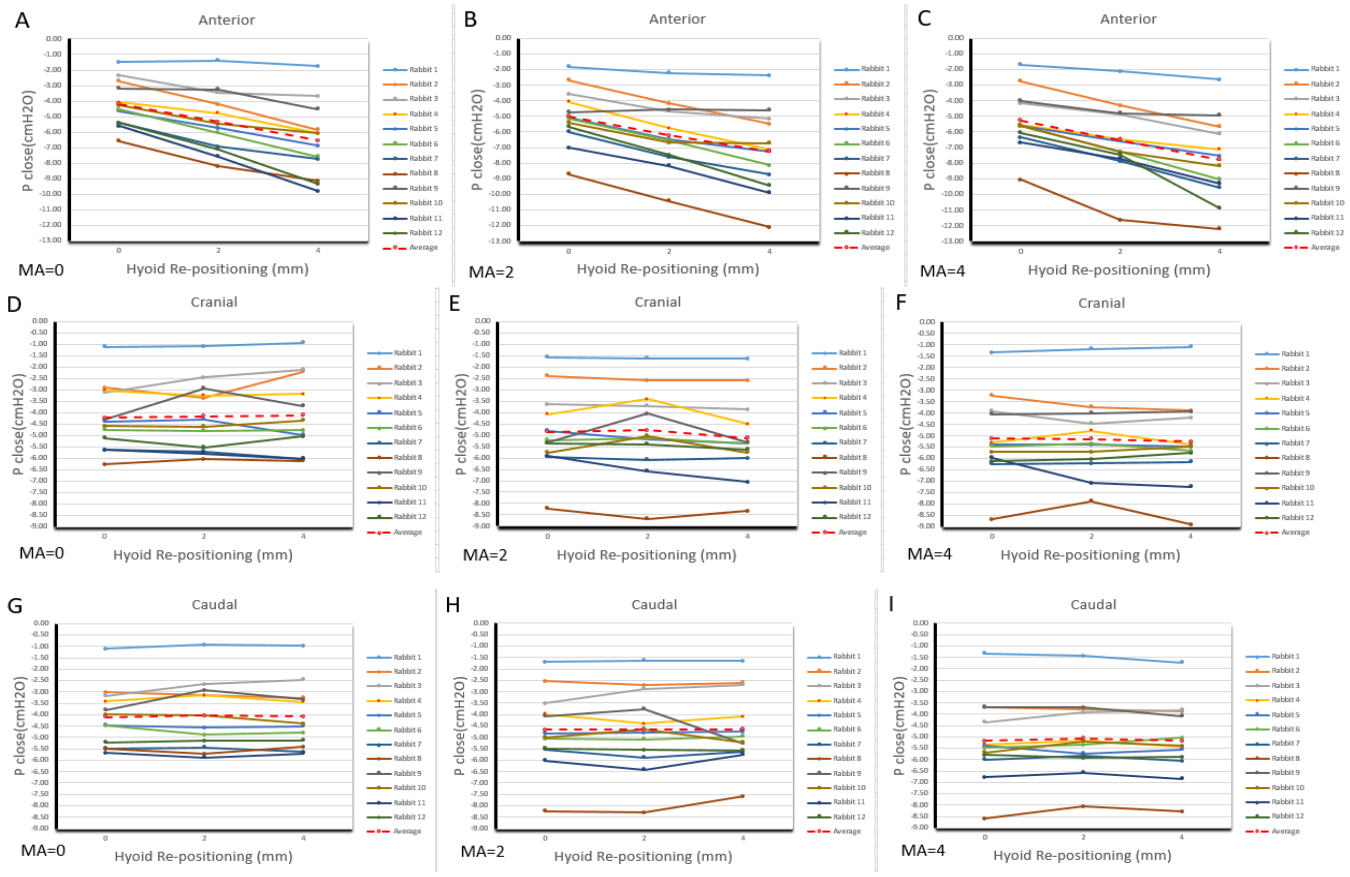


Figure IV 1: Graphs representing the absolute Pclose obtained in the 12 rabbits (represented seperately) with different mandibular advancement increment MA=0,2,4mm for various hyoid repositioning, from 0 to 2 and 4 mm, anterior (A:MA=0mm, B:MA=2mm, C:MA=4mm), cranial (D:MA=0mm, E:MA=2mm, F:MA=4mm) and caudal (G:MA=0mm, H:MA=2mm, I:MA=4mm). Each study is represented by a solid line and with a different color. The dashed line refers to the average Pclose in each direction for all the rabbits.

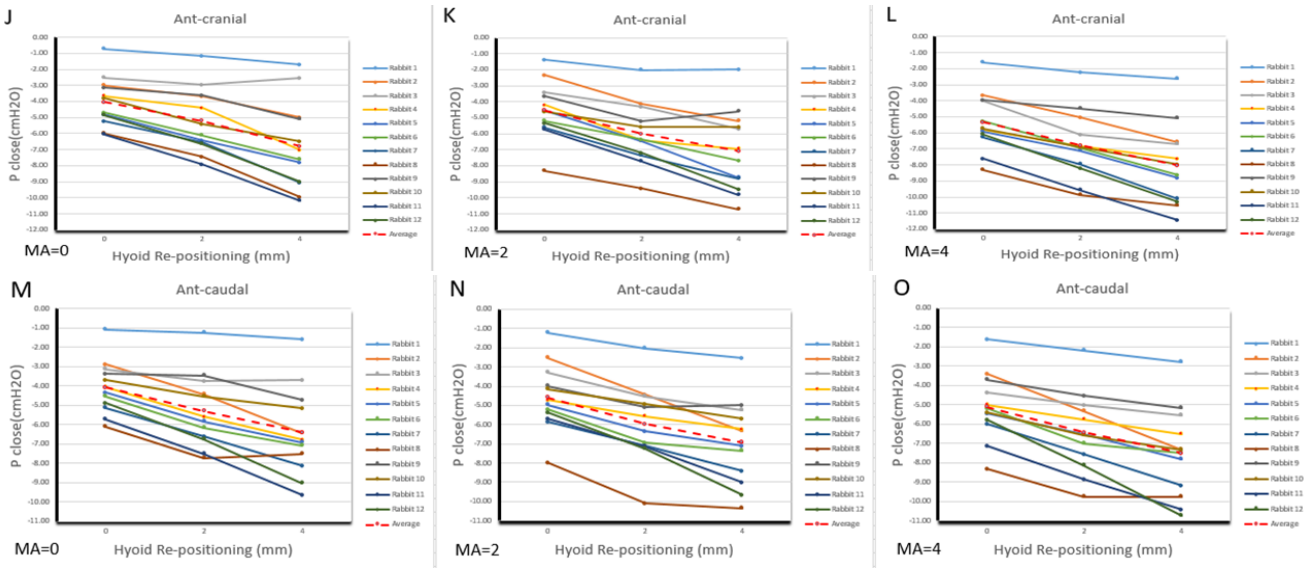


Figure IV 2: Graphs representing the absolute Pclose obtained in the 12 rabbits (represented seperately) with different mandibular advancement increment MA=0,2,4mm for various hyoid repositioning, from 0 to 2 and 4 mm, ant-cranial 45° (J:MA=0mm, K:MA=2mm, L:MA=4mm) and ant-caudal 45° (M:MA=0mm, N:MA=2mm, O:MA=4mm). Each study is represented by a solid line and with a different color. The dashed line refers to the average Pclose in each direction for all the rabbits.

D. Mandibular advancement

Highly statistically significant differences were found in absolute Pclose (p value=0.000) between the mandibular advancements of 0 and 2mm, 0 and 4mm and 2 and 4mm (table IV.3). When the increment of mandibular displacement was increased, Pclose decreased progressively. The Pclose values obtained with 0 and 2mm mandibular advancement were significantly higher at all incremental levels compared to 4mm of mandibular advancement. With no hyoid repositioning, the average absolute Pclose value obtained with MA=0mm was -4.1 cmH2O, compared with -5 cmH2O at MA=2mm, and -5.2 cmH2O at MA=4mm (Figure IV.2 A, D, G, J, M).

Table IV 3: Pairwise comparisons to assess which pairs of conditions significantly differ from one another for mandibular advancement increments

Pairwise Comparisons

Measure: Pclose

(I) MA	(J) MA	Mean Difference (I-J)	Std. Error	Sig.[b]	95% Confidence Interval for Difference[b]	
					Lower Bound	Upper Bound
0	2	.756*	.099	0.000	.476	1.036
	4	1.490*	.187	0.000	.963	2.017
2	0	-.756*	.099	0.000	-1.036	-.476
	4	.734*	.101	0.000	.450	1.019
4	0	-1.490*	.187	0.000	-2.017	-.963
	2	-.734*	.101	0.000	-1.019	-.450

Based on estimated marginal means

*. The mean difference is significant at the .05 level.

b. Adjustment for multiple comparisons: Bonferroni.

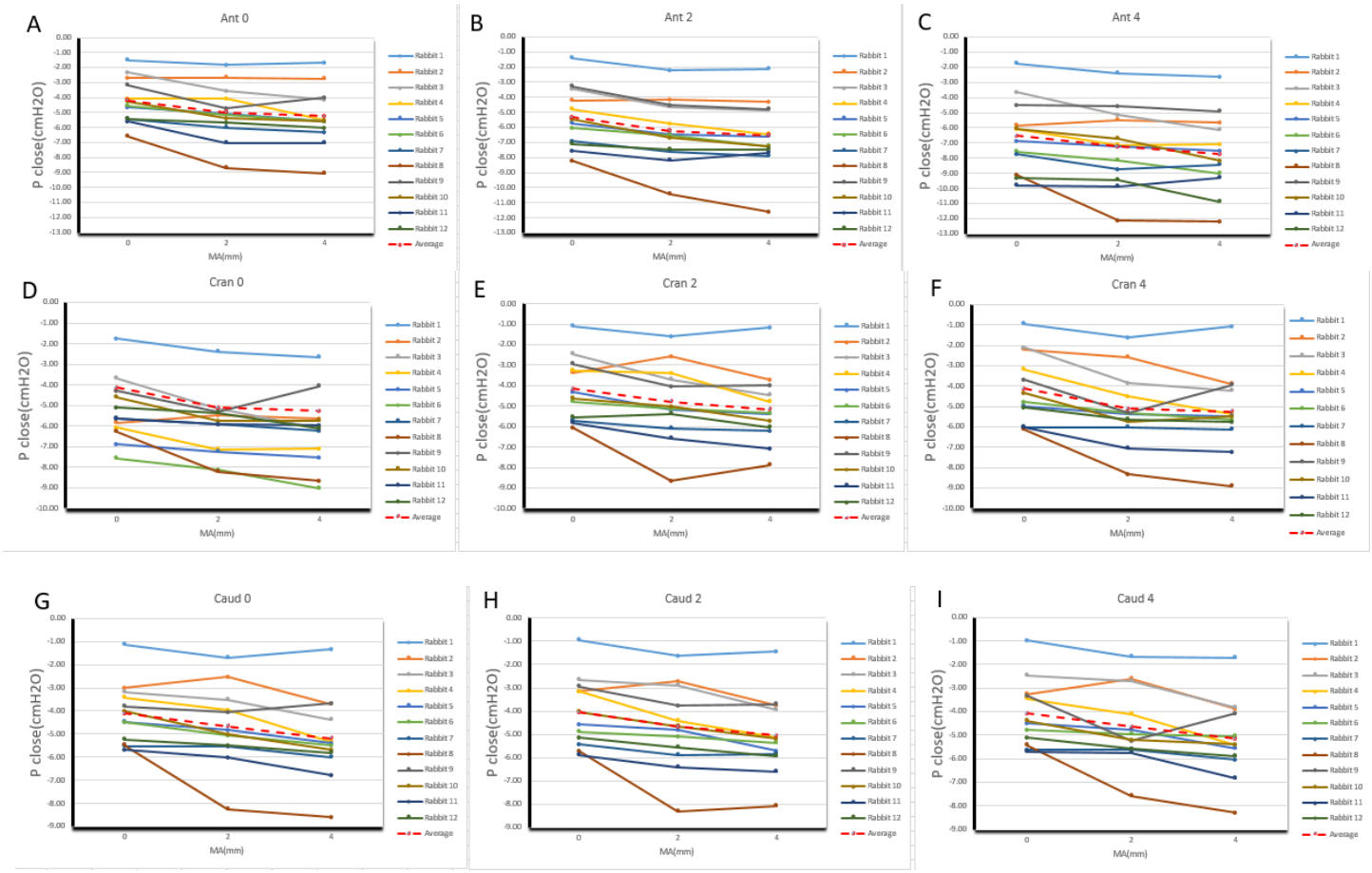


Figure IV 3: Graphs representing absolute Pclose values obtained in the 12 rabbits (represented seperately) for each repositioning direction, from 0 to 2 and 4 mm, anterior (A:0mm, B:2mm, C:4mm), cranial (D:0mm, E:2mm, F:4mm) and caudal (G:0mm, H:2mm, I:4mm) with different mandibular advancement increments (MA=0,2,4mm). Each study is represented by a solid line and with a different color. The dashed line refers to the average Pclose in each direction for all the rabbits.

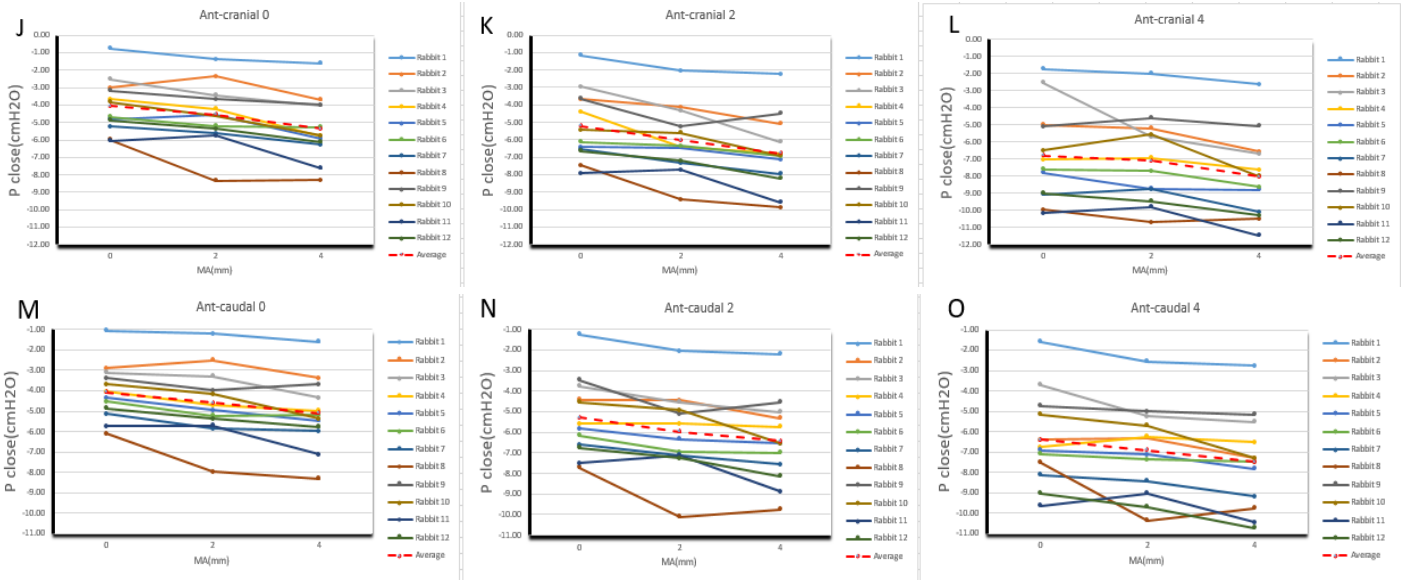


Figure IV 4: Graphs representing absolute Pclose values obtained in the 12 rabbits (represented seperately) for each repositioning direction, from 0 to 2 and 4 mm, ant-cranial 45° (J:0mm, K:2mm, L:4mm) and ant-caudal 45° (M:0mm, N:2mm, O:4mm) with different mandibular advancement increments (MA=0,2,4mm). Each study is represented by a solid line and with a different color. The dashed line refers to the average Pclose in each direction for all the rabbits.

E. Mandibular advancement and hyoid repositioning direction and increment combination

When MA was combined with hyoid repositioning, the average absolute P close was more negative. This was specifically observed with the anterior hyoid repositioning component (anterior, ant-cranial 45°, ant-caudal 45°) concomitant with gradual MA. The average absolute P close decreased from -4.9 cmH2O (when both mandible and hyoid were not repositioned) to -6 cmH2O with MA=2mm and hyoid anterior repositioning of 2mm, and to -7.1 cmH2O with MA=4mm and hyoid anterior repositioning of 4mm (Figure IV.3 A). The average absolute P close decreased from -4.6 cmH2O (when the bones were not repositioned) to -6 cmH2O with MA=2mm and hyoid ant-cranial repositioning of 2mm and to -7.4 cmH2O with MA=4mm and hyoid

ant-cranial repositioning of 4mm (Figure IV.3 D). The average absolute P close decreased from the initial -4.6 cmH2O to -6 cmH2O with MA=2mm and hyoid ant-caudal repositioning of 2mm and to -7 cmH2O with MA=4mm and hyoid ant-caudal repositioning of 4mm (Figure IV.3 E). However, when MA was combined with cranial and caudal hyoid repositioning, P close remained almost the same. The average absolute P close is of -4.7 cmH2O (when both mandible and hyoid were not repositioned) remained the same with MA=2mm and hyoid cranial repositioning of 2mm but decreased slightly to -4.9 cmH2O with MA=4mm and hyoid cranial repositioning of 4mm (Figure IV.3 B). For caudal hyoid repositioning, the average absolute P close value remained unchanged with various hyoid increments (0,2 and 4mm) and in concomitance with MA; the average absolute P close was -4.6 cm H2O (Figure IV.3 C).

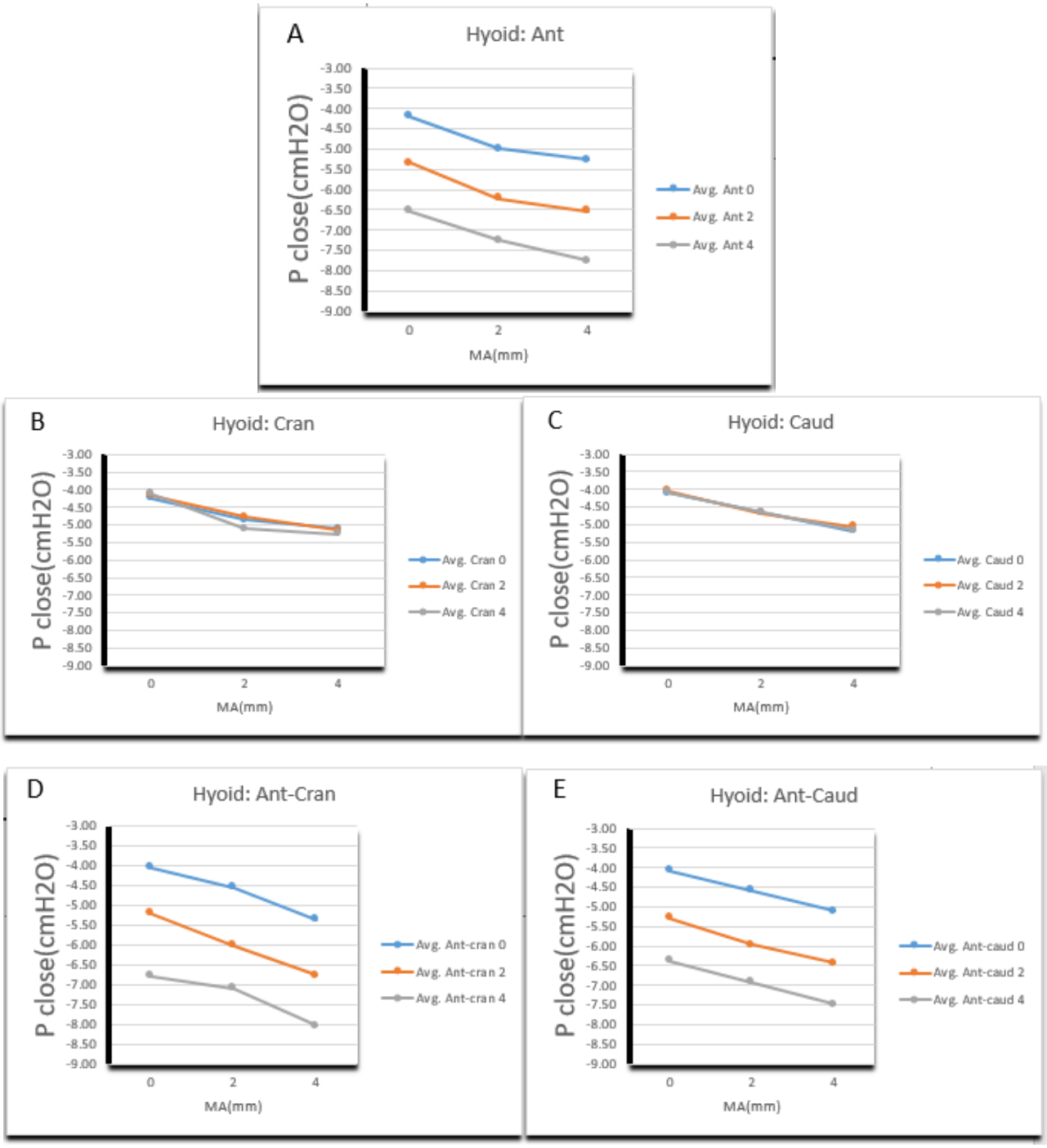


Figure IV 5: Graph representing the averages combined (of all 12 rabbits) absolute Pclose obtained for various hyoid repositioning directions from 0 to 2 and 4 mm (Graph A:anterior, B: cranial, C: caudal, D:ant-cranial 45°, E: ant-caudal 45°) with different mandibular advancement increments (MA=0,2,4mm).

CHAPTER V

DISCUSSION

This is the first study to investigate the effect of hyoid bone repositioning combined with mandibular advancement on upper airway collapsibility. Although the literature suggests a potential role for the hyoid bone and mandibular position to affect upper airway patency, no prior study has quantitatively assessed this effect.

The primary outcomes of this study were that:

1) Incremental anterior component repositioning of the hyoid bone alone leads to decrease in upper airway collapsibility.
2) Incremental mandibular advancement alone leads to decrease in upper airway collapsibility.
3) Anterior repositioning of the hyoid bone combined with mandibular advancement are the main factors leading to decreased upper airway collapsibility. This outcome is positively correlated with the range of displacement: the more the hyoid bone and the mandible were repositioned anteriorly the less collapsible the upper airway became.

A. Critique of methods

Rabbit model

The rabbit model was chosen because of its favourable upper airway anatomy that resembles the human airway. In particular, the rabbit has a mobile hyoid bone, which is the site of insertion of several muscles and ligaments that control its position, similar to the human hyoid bone [250]. In rodents, dogs, and cats the hyoid bone is

firmly attached by cartilage to the styloid process and thyroid cartilage, making the airway less collapsible. The anaesthetized rabbit passive upper airway model has been employed extensively in previous studies of upper airway mechanics, including those that established the mechanical effects of upper airway dilator muscles on upper airway respiratory activity [251-254], tracheal traction effects on upper airway in rabbits[49], sciatic nerve stimulation and its effects on upper airway resistance[255], effect of mandibular advancement on upper airway lumen size and tissue pressure [105, 221]. The outcomes of these studies have repeatedly shown a high level of correlation with the outcomes of human studies [250] . Invasive interventions cannot be applied in humans; thus the rabbit became an ideal model for upper airway study.

Passive upper airway

The anesthetized and isolated upper airway rabbit model was used to study the passive upper airway, i.e. with no upper airway muscle activity or airflow. The respiratory upper airway pressure reflexes were eliminated after isolation of upper airways. Anaesthetic infusion decreases upper airway muscle activity and leads to partial airway collapse, simulating the effect of sleep on the upper airway [256]. In the current study, we used ketamine and xylazine for anaesthesia. The use of ketamine avoids the central suppressant effects produced by barbiturate anaesthesia and maintains airway patency [257]. However, ketamine and xylazine promote salivary secretion in both animals and humans that might stimulate muscle contraction [258]. Consequently, the trachea was suctioned to avoid excessive secretions from swallow reflexes which might have altered our results and prevented the maintenance of a patent upper airway in the rabbit during the surgical intervention.

Measuring Pclose

A used reproducible measure for airway collapsibility in animal and human studies is the upper airway closing pressure (Pclose) [29, 31, 55, 207, 259-261]. We used Pclose to evaluate the effect of different hyoid and mandibular positions on upper airway patency. The measurement method that we used in our study is similar to that previously described in dogs [262] and rabbits [252, 253]. However, the experimental application of negative pressure in the trachea for Pclose measurement can stimulate reflexes and can introduce slight muscular activity. Nonetheless, Pclose remains an accepted measure of upper airway passive collapsibility specially that the results obtained are consistent, further supporting the use of this metric.

Tissue stretch and deformation

After several negative pressure applications to the upper airway which is composed of highly deformable tissue, a stretch phenomenon of those tissues can occur leading to a change in tissue elastance and consequently to collapse [253]. Therefore, we waited around 30 to 40 seconds between each measurement for the tissue to accommodate the stretch and return to its original state [95]. Nevertheless, the stretch of the upper airway soft tissues might be a reason why the absolute values of Pclose slightly changed between runs. However, the change in Pclose with each intervention remained consistent.

Upper airway secretions

In addition to potential secretions from the anaesthetic agents, the repeated application of negative pressure to the upper airway can result in pharyngeal secretions (fluids in the upper airway, saliva, and mucus). This type of secretion increases the surface tension in the upper airway mucosal lining and thereafter increases collapsibility

[252]. The secretion also stimulates the swallow reflexes which can affect the measurements. To avoid excess build-up of secretions, we cleared the upper airway between runs with suction (using a syringe and a small diameter non-compliant tube). This method was an efficient way to eliminate the pharyngeal fluids [29, 263].

Hyoid repositioning device

The hyoid repositioning device was custom designed in house for the current study. It was attached via a clamp to a mini screw inserted in the hyoid bone. Stability of this connection was determined visually (making sure the hyoid bone baseline position is maintained after clamping it to the device). In addition, a marking on the clamp and the screw were placed and used as landmarks to ensure that no slipping occurred when the hyoid bone was moved from 0 to 4mm anteriorly, cranially, and caudally. No rotation or translation movement was observed between the clamp and the mini screw while moving the hyoid bone and the connection was stable. The hyoid device was clamped onto the hyoid bone at the beginning of the study and it was not unclamped until the end of the surgery. All measurements were performed with the same original clamping.

The hyoid bone was fixed in the new position and unable to move with any additional load, i.e. mandibular advancement. This set-up is similar to hyoid repositioning surgeries, in which the hyoid bone is fixed to the mandible or the thyroid cartilage. During normal functioning (swallowing, speech, breathing), the hyoid moves in response to various active and passive loads. Experimentally, preserving the hyoid mobility after displacement is not feasible. However, additional research (i.e. with

computational modelling) should be undertaken in the future to determine if hyoid movement post repositioning is important for upper airway function.

Mandibular advancement device

The mandibular advancement device was custom designed in house for the current study using an acrylic mould and screw system. A slight amount of mouth opening was inevitable due to the thickness of acrylic material interfering between maxillary and mandibular teeth. Slight rotation of the mandible in clockwise rotation can also occur as a consequence of mouth opening. Mouth opening and mandibular rotation can affect and modify the size of upper airway after changing the vertical position of the mandible. Consequently, trimming of the maxillary and mandibular incisors was undertaken for each rabbit to control the amount of mouth opening. Glass ionomer material was used to cement the maxillary and mandibular teeth to the device. After cementation, a few minutes were needed for the luting material to set fully for better stability of the device.

B. Discussion of the results

Hyoid repositioning

Anterior hyoid repositioning directions decreased upper airway collapsibility and improved its stability and patency. The more the hyoid was repositioned anteriorly, ant-cranially 45° and ant-caudally 45°, the better the outcome on Pclose. Advancement of the hyoid bone has long been recognized to offer potential benefit in the treatment of OSA. The outcome of anterior hyoid repositioning was previously examined in human cadavers. They found that anterior hyoid advancement by 1cm produced airway opening

and improvement in upper airway airflow by almost 4 fold (to 3.83 ± 1.93 L/min) [264]. Previous studies showed an increase in inspiratory flow and a decrease in the negative trans-upper airway pressure swing in inspiration and expiration [103]. Moreover, the effect of anterior hyoid repositioning was also examined on anesthetized rabbits in previous studies including a recent one (2020)[265]. The findings are consistent with our results showing a reduction of Pclose whenever the hyoid bone was displaced anteriorly. This reduction reflects a more stable and less collapsible upper airway. This improvement varied with the range of hyoid displacement from 0 to 4mm; the greater the hyoid bone was displaced anteriorly, the more Pclose was negative and the more stable was the upper airway. Ant-caudal and ant-cranial hyoid displacement studies have been performed by our group [265]. However, previous studies only included muscle stimulation (supra and infra-hyoid muscles) which caused hyoid bone movement in antero-cranial and antero-caudal directions. Based on our study and the previous study done by Samaha and Amatoury [265], the ant-caudal and ant-cranial movement improved the upper airway patency and stability. The improvement obtained is similar to the one obtained with anterior displacement of the hyoid bone. However, the caudal and cranial directions did not have any significant effect on upper airway closing pressure at all repositioning increments (0,2 and 4mm). Moreover, the Pclose was significantly higher at all incremental levels in comparison to Pclose obtained with the anterior component displacement (anterior, ant-cranial 45 ° and ant-caudal 45°).

Mandibular advancement

Incremental mandibular advancement induced a reduction in P close affecting the upper airway positively by decreasing upper airway collapsibility. P close is more

negative when the mandible is advanced by 4mm compared to 0 and 2mm. Previous findings suggested that MA increases pharyngeal patency by reducing airway resistance in anesthetized or awake individuals. Consequently, upper airway becomes less collapsible [219]. In fact, incremental mandibular advancement reduces upper airway length and enlarges upper airway cross sectional area and tissue stress [221]. It is also conceivable that mandibular advancement could be indirectly targeting the hyoid bone, which in turn is directly responsible for airway dilatation [28]. As the mandible is advanced, the hyoid, through its muscular connections to the body of the mandible, is displaced in an ant-cranial direction. This movement was found to improve the upper airway patency and stability [107]. However, in our study the hyoid was fixed in a specific position even after repositioning due to the attachment using a clamp. This method proves that even without the indirect effect of displacement of the hyoid after MA, MA will always improve upper airway collapsibility. This contrasts conceptual analysis that the ability of MA to enlarge the upper airway may be impaired if hyoid bone movement is restricted because MA increases upper airway lumen size. Greater improvement in Pclose could be observed if hyoid could move, because the hyoid bone plays an important role in redistributing the mandibular advancement load throughout the peripharyngeal tissue mass [266]. A computational finite element model of the rabbit upper airway was studied [267]. They found that for mandibular advancement, tissue displacements were reduced in the tongue and tissue mass with the hyoid bone fixed, compared with when the hyoid bone was free to move. Also minimal displacements and stresses/strains developed in tissues caudal to the hyoid bone [267].

Combined hyoid repositioning and mandibular advancement

Incremental anterior hyoid repositioning alone decreased P close, incremental MA alone also decreased P close. However, when MA was combined with anterior based hyoid repositioning there was a further decrease in P close. P close decreased from -5.2 cmH2O with MA=4mm and hyoid=0 to -7.8 cmH2O with MA=4mm and hyoid ant=4mm (50% improvement after hyoid displacement relative to no displacement) and from -5.2 cmH2O for MA=4mm and hyoid=0 to -8 cmH2O with MA=4mm and hyoid ant-cranial=4mm (53.84% improvement). Moreover, P close decreased from -5 cmH2O with MA=4mm and hyoid=0 to -7.4 cmH2O with MA=4mm and hyoid ant-caudal=4mm (48% improvement). Pclose obtained with MA=4mm and hyoid ant-cranial 45° repositioning of 4mm provides the best outcome and the greatest decrease in collapsibility with a Pclose of -8cmH2O which is equivalent to 53.84% of collapsibility improvement. This combination gives the best outcome because tissues including upper airway muscles are all being stretched in the same direction. As previously concluded by Amatoury et al [266], 1mm of MA induced on average 0.42mm of ant-cranial hyoid displacement. Combining MA with further hyoid ant-cranial repositioning contributes to UA improvements via greater redistribution of peripharyngeal tissue stresses [266]. This combined treatment approach might be of great advantage in improving airway patency. In a systematic review, Song et al [268] have shown that in patients undergoing isolated hyoid surgery for OSA, hyoid suspension could reduce the apnea-hypopnea index (AHI) by only 38.3%. Another study concluded that hyoid suspension alone is not an efficacious treatment for obstruction in OSA for most patients[269]. Therefore, the literature is still not

conclusive on the efficacy of the hyoid suspension as a part of a combined therapeutic approach with mandibular advancement. Other studies specified that in awake OSA subjects larger increments of hyoid bone displacement with mandibular advancement are linked to greater improvements in upper airway lumen size [107]. However, no quantitative studies revealed the real interaction between MA and hyoid repositioning. Information on the behavior of upper airway after a combination of hyoid and mandibular protrusion is both meagre and diverse, and the results of this maneuver on pharyngeal opening were unresolved. This study was needed to understand the physiological changes following mandibular advancement therapy and the relation to the hyoid bone. Upper airway patency and stability improved after combining hyoid and mandibular anterior displacement, and this improvement was dose dependent (Figure V.1). These findings suggest that hyoid repositionning surgeries should aim at maximizing the anterior hyoid displacement and they should be combined with mandibular advancement using a splint to reposition the mandible anteriorly by avoiding excessive advancement and tissue stretch, damage and potential side effects. Moreover, hyoid repositioning surgeries can be combined with surgical mandibular advancements, but this approach needs further studies. In fact, hyoid surgeries are difficult to apply clinically specially that mandibular advancement alone is effective in displacing the hyoid cranially. This elucidates that surgical or non-surgical mandibular advancement can induce hyoid displacement without the need to do any surgery at the level of the hyoid bone. However, the question is if the amplitude of hyoid displacement following mandibular advancement alone is enough or not. The hyoid could be displaced more cranially as a consequence of surgical mandibular advancement compared to non-surgical mandibular advancement using a splint. In addition, the

advantage of surgical mandibular advancement is that the mandible can be moved anteriorly but also cranially, in contract to mandibular advancement using a splint that leads to the repositioning of the mandible anteriorly but caudally. All these differences can be assesed by comparing MAS to MMA and the effect of these two interventions on hyoid position and the combination of these intervention and their effect on upper airway patency.

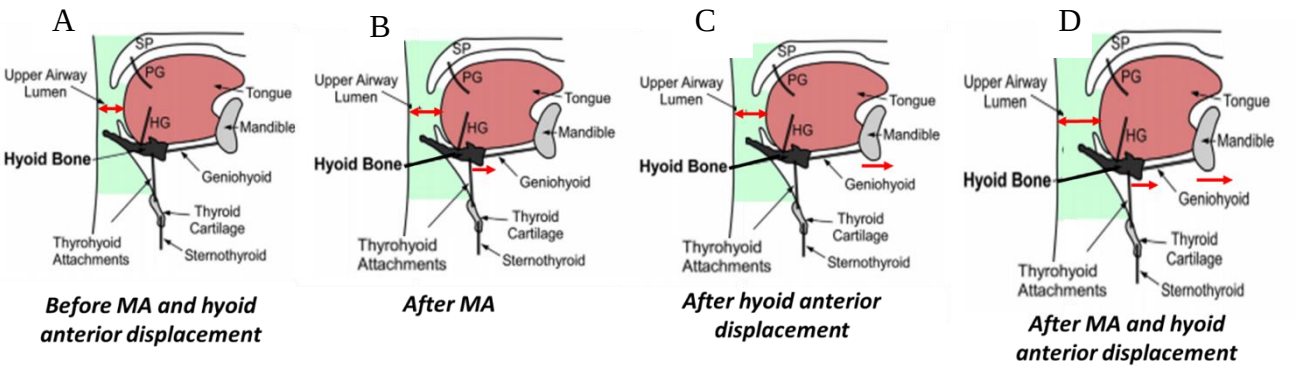


Figure V 1. Schematic description of the upper airway at baseline (A). After MA (B). After anterior hyoid displacement (C) and after MA and hyoid anterior displacement combined together (D).

CHAPTER VI

CONCLUSION AND CLINICAL IMPLICATIONS

1- The incremental increase of hyoid bone repositioning was correlated with a progressive reduction in the upper airway closing pressure leading to a less collapsible upper airway. This improvement was observed in the anterior and ant-cranial/ ant-caudal 45° angles. The incremental increase of mandibular advancement was also correlated with a progressive reduction in the upper airway closing pressure leading to a less collapsible upper airway.
2- When mandibular advancement was combined with hyoid reposition in anterior based directions, upper airway closing pressure was reduced further leading to a decreased collapsibility of upper airway.

This study suggests that for OSA patients where MA alone is not successful in treating their OSA, MA combined with anterior based hyoid repositioning may help in treating OSA.

During mandibular advancement therapy, the hyoid bone moves in an antero-cranial direction which was proven to improve the upper airway patency. Increasing the amount of mandibular advancement (surgical or using a repositionning device) tends to increase the anterior displacement of the hyoid bone (following the force vectors). Therefore, different ranges of displacement of hyoid bone with mandibular advancement may explain differences in the pattern of improvement in upper airway collapsibility between response groups in previous studies [36, 270]. Clinically, the practitioner should create a balance between the patient's comfort and maximum mandibular advancement.

This study helps in determining the relation between hyoid bone position and the efficacy of mandibular advancement therapy and how hyoid position influences mandibular advancement to modify upper airway patency. This prospects provides fundamental evidence in understanding the hyoid bone's role in the efficacy of a primary 2nd line therapy for OSA, which is currently only 50% successful. Since the efficacy of MA on Pclose and airway collabsibility is proven, MA can be considered as a 1st line therapy for patients with mild to moderate OSA.

A. Future Studies

The findings of this study are based on an animal model which has a highly similar upper airway structure to humans. Nevertheless, it would be ideal if those interventions would be translated to humans, to study the species we are interested in with this disorder. But that is not possible due to invasive procedures.

Comparative studies of surgical and non-surgical mandibular advancements and the effect on hyoid position should be assessed. Moreover, the effect of the combination of these interventions on upper airway collapsibility should be examined.

For an optimal physiological representation, a great addition to this study would be to include imaging through a lateral cephalometric radiographic examination to assess dimensional changes at the level of the airways after mandibular and hyoid repositioning. Such investigation will allow to examine the exact changes, obtained at different airway levels, before and after each intervention and determine how these interventions are modifying Pclose for targeted therapy. The changes will be better understood at the tissue level in terms of pressure and strain.

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