ACTA Otorhinolaryngologica Italica

Official Journal of the Italian Society of Otorhinolaryngology Head and Neck Surgery

Organo Ufficiale della Società Italiana di Otorinolaringoiatria e Chirurgia Cervico-Facciale

S.I.O.E.C.I.C.-F

PACINI EDITORE MEDICINA

Volume 39
4
August 2019

www.actaitalica.it

Acta Otorhinolaryngologica Italica

Official Journal of the Italian Society of Otorhinolaryngology Head and Neck Surgery

Organo Ufficiale della Società Italiana di Otorinolaringoiatria e Chirurgia Cervico-Facciale

Managing Editor
M. Ansarin

Publisher
Pacini Editore Srl
Via Gherardesca, 1 - 56121 Pisa, Italy
Tel. +39 050 313011 -Fax +39 050 3130300
info@pacinieditore.it - www.pacinimedicina.it

Acta Otorhinolaryngologica Italica is cited in Index Medicus, MEDLINE, PubMed Central, Science Citation Index Expanded, Scopus, Open-J Gate, Free Medical Journals, Index Copernicus, Socolar

2018 Journal Impact Factor, Journal Citation Reports (Web of Science Group, 2019): 1.408
Acta Otorhinolaryngologica Italica is available on Google Scholar

S.I.O.E.C.I.C.-F

PACINI EDITORE MEDICINA

Volume 39
August 2019
4

www.actaitalica.it

Contents

Articles in open access, information for authors including editorial standards for the preparation of manuscripts and submission and Publisher's note available on-line: www.actaitalica.it

ACTA OTORHINOLARYNGOLOGICA ITALICA 2019;39:211-219; doi: 10.14639/0392-100X-2155

Review

Management of idiopathic epistaxis in adults: what's new?

Il trattamento dell'epistassi idiopatica nell'adulto: cosa c'è di nuovo?

G. MECCARIELLO[1], C. GEORGALAS[2], F. MONTEVECCHI[1], G. CAMMAROTO[1 3], R. GOBBI[1], E. FIRINU[1], A. DE VITO[1], C. VICINI[1]

[1] Department of Head-Neck Surgery, Otolaryngology, Head-Neck and Oral Surgery Unit, Morgagni Pierantoni Hospital, Forlì, Italy; [2] Department of Otolaryngology, Hygeia Hospital, Athens, Greece & Leiden University Hospital, Leiden, The Netherlands; [3] Department of Otolaryngology, University of Messina, Italy

SUMMARY

Epistaxis is one of the most common complaints presenting to emergency departments. The aim of this study is to systematically review and critically evaluate the evidence relating to treatment of idiopathic epistaxis for guiding best practice. A comprehensive review of the English language literature was performed using PubMed, Embase, Cochrane Library and Central electronic databases. The inclusion criteria were: retrospective or prospective or randomised controlled clinical trials which included outcomes in the management of idiopathic epistaxis. Twenty-three articles met inclusion criteria and were reviewed. Nasal packing still represents the first-line approach to epistaxis, although, at present, it appears that there is clear evidence in the literature to suggest that it is less effective and associated with more admissions and longer hospital stays than endoscopic electrocoagulation-based management of epistaxis. In conclusion, cauterisation should be the first-line approach for its high cost-effectiveness rate and low risk of complications. Further research is urgently needed to assess the efficacy of new biomaterials.

KEY WORDS: Epistaxis • Protocol • Endoscopy • Cautery • Sphenopalatine artery • Embolisation

RIASSUNTO

L'epistassi è uno dei disturbi più comuni per il quale il paziente si rivolge spesso al pronto soccorso. Questa revisione della letteratura si propone di valutare sistematicamente e criticamente gli studi scientifici riguardo il trattamento dell'epistassi idiopatica al fine di ottenere utili spunti per la pratica clinica. La ricerca è stata eseguita nei database elettronici: PubMed, Embase, Cochrane e Central. I criteri di inclusione sono stati: studi clinici controllati retrospettivi o prospettici o randomizzati o studi su modelli animali che includevano i risultati nella gestione dell'epistassi idiopatica. Sono stati individuati 23 articoli che soddisfano i criteri di inclusione. Il tamponamento nasale rappresenta ancora l'approccio di prima linea all'epistassi, anche se è evidente dalla letteratura che sia il meno efficace ma il più associato a ricoveri ospedalieri di maggior durata rispetto alla chirurgia endoscopica basata sull'elettrocoagulazione. In conclusione appare sempre più evidente che la cauterizzazione dovrebbe essere l'approccio di prima linea per l'alto tasso di costo-efficacia e il basso rischio di complicanze. Tuttavia, ulteriori ricerche urgenti sono necessarie per validare l'efficacia dei nuovi biomateriali nel trattamento dell'epistassi.

PAROLE CHIAVE: *Epistassi • Endoscopia nasale • Cauterizzazione • Arteria sfenopalatina • Embolizzazione*

Introduction

Epistaxis is one of the most common presenting symptom to both primary care and accident and emergency departments. It is thought to affect 10-12% of the population [1], and although most cases are self-limiting, some do not resolve without intervention. Among these, 80% are from the Kiesselbach's plexus (anterior epistaxis) [2]. Interestingly, some studies have suggested a positive correlation between epistaxis and atmospheric pressure or relative humidity and changes in temperature [3 4]; others also included allergic rhinitis as one of the main causes of epistaxis [5]. Only 6% of patients with epistaxis will require medical assistance, and cases of severe intractable epistaxis are rare [6]. Anterior nasal packing, the most common therapy for epistaxis, has some limitations including potential for reduced ventilation and sleep apnoea, need for analgesics and, in some cases, need for prophylactic antibiotics [6]. Nasal cautery forms an important part of first line management, and is considered an important skill for anyone treating epistaxis [7], but is unlikely to be practiced by non-otolaryngologists. The goal of this study was to assess current trends in the management of epistaxis in the adult population.

The aim of this study is to systematically review and critically evaluate the evidence relating to treatment of idiopathic epistaxis in order to guide best practice.

Search strategy

A comprehensive review of the English language literature was performed using PubMed, Embase, Cochrane Library and Central electronic databases using the keyword "epistaxis". The search was executed in March 2016 and was limited exclusively to studies published after 2010. It yielded a total of 1917 articles. We retrieved the full text of any reports potentially meeting inclusion criteria and examined these independently to determine study eligibility. Articles were examined for data resolution with the intent to perform a meta-analysis. Different methods of meta-analyses were considered in reviewing the literature to seek results that would provide meaningful analysis with the least risk of introducing biases. The inclusion criteria were: retrospective or prospective or randomised controlled clinical trials which included outcomes in the management of idiopathic epistaxis. Exclusion criteria were case reports or letters/correspondence to editor; clinical studies including the management of epistaxis other than idiopathic or mixed cases in which was not possible to extrapolate data concerning idiopathic epistaxis; paediatric population or animal model; systematic reviews and meta-analyses; non-English language manuscripts. To reduce the risk of incomplete literature search, a manual search of the references of included papers was performed (Fig. 1 for flowchart). Data from studies were first extracted and assessed by the principal investigator (MG) and thereafter independently by 2 co-authors (MF and GC) using standardised data forms. Quality assessment of studies (QUADAS-2) tool was used to evaluate relevant study design characteristics of included studies [8]. A graphical display of QUADAS-2 results is shown in Figure 2.

Results

The search was performed in March 2016 and yielded 1,917 articles. Moreover, 2 records were added from a manual search. Twenty-three articles [9-31] met inclusion criteria. An overview of included studies is included in Table I.

Nasal packing

The first-line therapy includes anterior-posterior synthetic nasal packs and balloons in most studies [9 10 17 18 20 21 23 26 27 31 32 34 35]. More specifically, the effectiveness of chitosan-based packing was assessed by Kourelis and Shikani [24]. Chitosan is a natural cationic polysaccharide with well-known powerful haemostatic properties. The study group consisted of 35 consecutive patients with drug-induced bleeding diathesis. The pack consisted of a single sheet of ChitoFlex® (Hemcon, Inc. Portland, OR, USA), wrapped around a hydroxylated polyvinylacetal sponge (Pope Merocel®, Medtronic, Inc, Minneapolis, MN, Usa). The chitosan-based nasal packing achieved instant control of bleeding in 32 cases (91%). The mean time to bleeding cessation was 3.5 min, ranging from 1 to 10 min. Moreover, a comparison among different types of nasal packings was performed by Dutta et al. [17]. Three methods were included for the anterior nasal packing, nasal packing-merocel (group A), gauze pack lubricated by sisomicin ointment (group B) and the same gauze pack used with a splint made of a sterilised aluminium foil sheet prepared from the cover of the suture materials used over the septum (group C), carefully using the paper-covered surface of the foil to remain in contact with the mucosa to eliminate any systemic absorption of aluminium. In 26.7% (n = 64), patients nasal tampon was used (30 for epistaxis and 34 for postoperative pack); 39.2% (n = 94) of patients were treated with conventional gauze pack with sisomycin cream (60 for epistaxis and 34 for post operative pack). The remaining 34.2% (n = 82) of patients were treated with the gauze pack with splint as described earlier (42 for epistaxis and 40 for postoperative pack). The "Behavioural Observational Pain Rating Scale" measured perception of pain during the procedure of packing. Fifty-six of 60 patients (93.33%) with gauze packing for

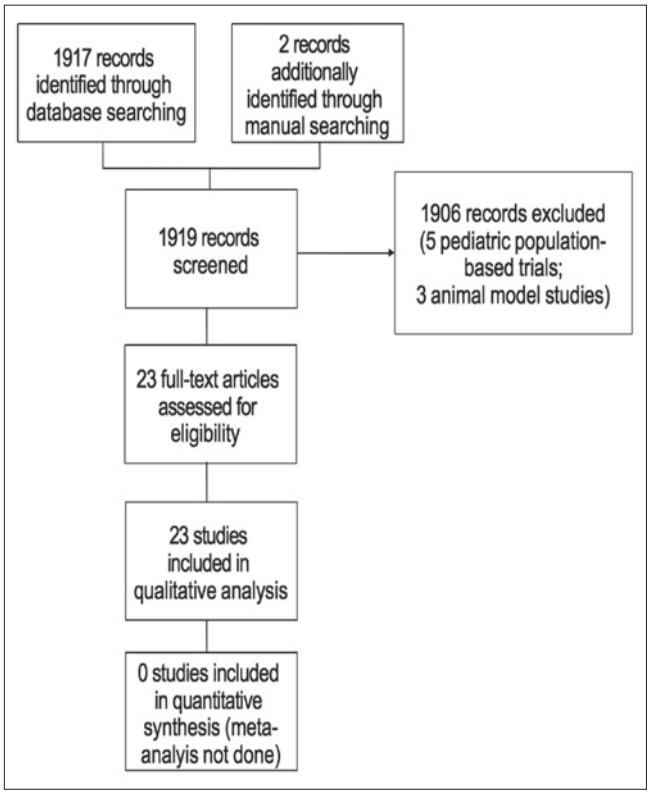

Fig. 1. Process for sifting search results and selecting studies for inclusion.

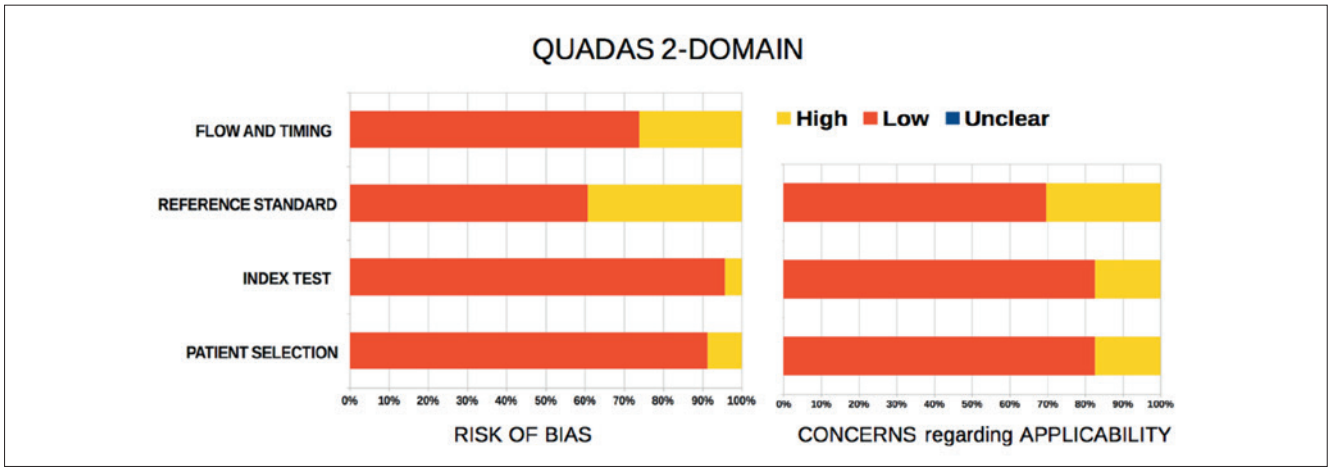

Fig. 2. Proportion of studies with unclear, low and high risk of bias and concerns regarding applicability.

Table I. Included studies.

Authors	Publication year	Study design	N of patients	Site of epistaxis	Materials	Outcomes
Lau et al. [9]	2016	Prospective, control-matched, longitudinal	20 (cases) 20 (control group)	Anterior and posterior	FloSeal vs nasal packing	FloSeal efficacy 75% vs nasal packing 85% (NS)
Zou et al. [10]	2015	Retrospective	53	Posterior fornix of the inferior nasal meatus	Endoscopic bipolar electric haemostat vs nasal packing	Lower VAS score, rate of re-bleeding, and nasal cavity adhesion for endoscopic bipolar electric haemostat ($p < 0.001$)
Limura et al. [11]	2015	Retrospective	167	Posterior	Electrocoagulation vs gauze tamponade vs balloon tamponade vs follow-up	Recurrent bleeding rates based were 8.5% (8/94) for electrocoagulation, 41.2% (21/51) for gauze tamponade, 50% (1/2) for balloon tamponade, and 45% (9/20) for follow-up
Butrymowicz et al. [12]	2015	Anatomical model and prospective case series	4	Anterior	Endoscopic electrocautery greater palatine artery	No recurrences
Khan et al. [13]	2015	Prospective	101	Anterior and posterior	FloSeal vs nasal packing	Success rate: 14%
Kilty et al. [21]	2014	Prospective	20	Posterior	FloSeal	Success rate: 80%
Shrestha [14]	2014	Retrospective	12	Posterior	Endoscopic sphenopalatine ligation	Success rate: 100%
Henderson et al. [15]	2013	Retrospective	124	Anterior and posterior	Electrocautery vs nasal packing	The rates of admission and of nasal packing reduced with the increasing of usage of electrocautery ($p < 0.001$)
Gandomi et al. [16]	2013	Retrospective	27	Posterior	Endoscopic sphenopalatine artery ligation	Success rate 87%
Gottumukkala et a.l [18]	2013	Retrospective	84	Posterior	Embolisation of one or more external carotid artery branches	Success rate of 89%
Zahed et al. [19]	2013	Randomised controlled	109 (anterior nasal packing group) 109 (topical tranexamic acid group)	Anterior	Anterior nasal packing vs topical tranexamic acid	Recurrence rates: 11% for anterior nasal packing, 2.8% for topical tranexamic acid ($p = 0.018$)
Shargorodsky et al [20]	2013	Retrospective	147	Anterior and posterior	Nasal packing vs silver nitrate vs dissolvable packing	Failure rate 23.8% for silver nitrate vs 57.4% for nasal packing vs 20% dissolvable packing

continues ▶

Table I. *follows.*

Authors	Publication year	Study design	N of patients	Site of epistaxis	Materials	Outcomes
Dutta et al. [17]	2012	Prospective	132	Anterior	Merocel vs gauze with sisomicin cream vs gauze with sisomicin cream and septal splint	No recurrence rates after removal: 96.9% for Merocel, 89.4% for gauze with sisomicin cream, 97.6% for gauze with sisomicin cream and septal splint (NS)
Mudunuri, Murthy [22]	2012	Prospective	42 (conservative group) 52 (silver nitrate group)	Anterior	Silver nitrate vs conservative	Failure rate: 26% for silver nitrate, 30% for conservative
George at al. [23]	2012	Retrospective	25	Anterior and posterior	Endoscopic sphenopalatine artery ligation	Success rate 88%.
Kourelis, Shikani [24]	2012	Prospective	35	Anterior	Chitosan-based packing	Success rate: 91%.
Baloch et al. [25]	2012	Retrospective	16	-	Angioembolisation	Success rate: 87.5%
Zhang and Qiu [26]	2012	Randomised controlled	41 (Nd:YAG Laser) 41 (liquid paraffin plus antiseptic cream)	Anterior	Nd:YAG laser vs liquid paraffin plus antiseptic cream	Success rate: 85% for Nd:YAG laser vs 40% for control group
Strach et al. [29]	2011	Retrospective	31	-	Endovascular embolisation with polyvinyl alcohol particles	Long-term success rate: 93.5%
Eladl et al. [28]	2011	Prospective	42	Posterior	Endoscopic sphenopalatine artery ligation	Success rate: 100%
Lesley et al. [27]	2010	Retrospective	20	Posterior	Angioembolisation with detachable platinum fibered coils	Success rate: 95%
Cotê et al. [30]	2010	Prospective	10	Posterior	FloSeal	Success rate: 80%
Minni et al. [31]	2010	Retrospective	48	Anterior and posterior	Endoscopic cautery of sphenopalatine artery or anterior ethmoidal artery	Success rate: 93%

VAS: visual analogue scale; NS: not statistical significant.

epistaxis experienced severe pain (score 8-10), while 36 of 42 (85.71%) experienced severe pain when aluminium foil splints were used. When nasal tampon was used, only 4 of 30 patients (13.33%) felt severe pain. This difference was statistically significant. Obviously, pain could not be recorded if the packing was inserted under general anaesthesia during septal surgery. With gauze pack in situ, 22 of 34 patients (64.70%) experienced moderate pain, in 6 patients (17.65%) pain was mild and another 6 patients felt severe pain. With the addition of septal splint, 10 of 40 patients (25%) felt mild pain, 28 (70%) had moderate pain and 2 patients (5%) experienced severe pain. Nasal tampon packing was almost painless in postoperative group as 30 of 34 patients (88.23%) felt only mild pain. The episodes of bleeding while packed in situ, within first 48 h and forced for repacking, was significantly more prevalent among nasal tampon group (group A) of patients, but as such no difference was revealed between groups B and C. Regarding experience of bleeding after removal of packing, no difference could be observed between nasal tampon and gauze packing with or without splint. Whereas group B showed a significantly higher rate of bleeding compared to group C. Synaechia formation was not lower among group A patients; however, comparison with the other two groups showed that the event was more common among subjects who belonged to group B vs group C, with a statistically significant difference.

Electrocoagulation

In other studies [9 10 17], the efficacy of nasal packing was compared to electrocoagulation. Zou et al. [10] compared the outcomes between a group of 15 patients who underwent nasal packing for inferior meatus epistaxis and a group of 38 patients who had electrocoagulation using bipolar forceps both under general and local anaesthesia. The incidence of re-bleeding was significantly lower in the electrocoagulation group (0/38) than in the packing group (4/15; $p = 0.001$). Re-bleeding occurred within 48 and 24 hours after nasal pack removal in three and one patient, respectively. All patients with re-bleeding underwent endoscopic surgery with successful achievement of haemostasis. The discomfort visual analogue score (VAS) differed significantly between groups, ranging from 1 to 5 in the surgery group (2.4 ± 1.4) and 6 to 9 in the packing group (7.6 ± 1.0; $p = 0.001$). The incidence of nasal cavity adhesions was significantly lower in the surgery group (2/38) than in the packing group (7/15; $p = 0.007$). Iimura et al. [11] evaluated 167 patients with idiopathic

posterior epistaxis. Bleeding sites included the olfactory cleft in 39 of 167 patients (23.4%), middle meatus region in 44 patients (26.3%), inferior meatus region in 36 patients (21.6%), other in 8 patients (4.8%) and unknown in 40 patients (24.0%). Electrocoagulation was performed in 94 patients (56.3%), gauze tamponade in 51 patients (30.5%), balloon tamponade in 2 patients (1.2%) and follow-up in 20 patients (12.0%). Of note, electrocoagulation was performed using an endoscope in patients in whom electrocoagulation under direct vision was not possible. Monopolar forceps were used on bleeding sites where haemostasis using bipolar forceps (straight/curved) was difficult. Recurrent bleeding occurred in 39 of 167 patients (23.4%). The bleeding sites at initial examination in 39 patients with recurrent bleeding were the olfactory cleft, middle meatus and inferior meatus, in 6, 8 and 4 patients, respectively, and was unknown in 21 patients. The recurrent bleeding rates based on bleeding site (patients with recurrent bleeding/patients with bleeding from the site) were 15.4% (6/39) for the olfactory cleft, 18.2% (8/44) for the middle meatus, 11.1% (4/36) for the inferior meatus and 52.5% (21/40) if the bleeding point was unknown. Haemostatic procedures at initial examination for 39 patients with recurrent bleeding were electrocoagulation (n = 8), gauze tamponade (n = 21), balloon tamponade (n = 1) and follow-up (n = 9). The recurrent bleeding rates based on the haemostatic procedure were 8.5% (8/94) for electrocoagulation, 41.2% (21/51) for gauze tamponade, 50% (1/2) for balloon tamponade and 45% (9/20) for follow-up, with electrocoagulation showing the lowest rate of recurrent bleeding.

Henderson et al. [15] assessed the benefit of implementing a standardised treatment protocol for adult epistaxis management based upon the use of bipolar electrocautery in preference to nasal packing. The study compared the results of epistaxis treatment before and after introduction of the protocol. There were 61 and 63 individuals included in the pre- and post-protocol groups. The introduction of bipolar electrocautery as a treatment modality into the post-protocol group resulted in a significant change in treatment outcomes. The frequency of nasal packing reduced significantly from 34 (56%) to 14 (22%) (p = 0.0002), with a corresponding rise in bipolar electrocautery from nil to 27 (43%) cases. The use of $AgNO_3$ remained relatively constant between both groups, despite not being on the intervention protocol. Admission rates fell significantly between the two groups from 38 to 23 (62-37%, p = 0.0068). Admissions for social/medical reasons rose very slightly in the post-protocol group with an increase in social admissions (5.7%). There were no cases recorded as readmissions. The overall average hospital length of stay was 1.2 days for the pre-protocol group, 0.83 days post-protocol and 2.4 days for those who underwent nasal packing in either group. Using actual numbers of overnight admissions, ward attenders and treatment used, total costs were £ 24,706 for the pre-protocol group, and £ 18,175 for the post-protocol group. This represented a per-patient average saving of £ 117, and per year £ 43,345 assuming an average of 31 presentations per month.

FloSeal

The usage of FloSeal haemostatic matrix (Baxter Healthcare, Deerfield, IL, USA), a human thrombin-impregnated, bovine gelatin matrix, has been evaluated in managing epistaxis alone [21 30] or compared to nasal packing [9 13]. Kilty et al. [21] evaluated this matrix in 20 patients with posterior epistaxis. Gelatin-thrombin matrix successfully stopped posterior epistaxis in 16 cases (80%). All of these cases received a single gelatin-thrombin matrix treatment. Four patients (20%) required additional treatment after gelatin-thrombin matrix failed to stop posterior epistaxis; two had surgical treatment; and two had posterior packing. No patient complications occurred in this study. Anticoagulant use was not significantly associated with treatment failure (p = 1.0). There was likewise no association with gender (p = 0.58), hypertension (p = 1.0), or diabetes (p = 0.62). VAS assessment for pain with the gelatin-thrombin matrix treatment had a mean of 3.6. On the other hand, Côté et al. [30] observed that epistaxis was adequately controlled in 8 of 10 patients without any adverse events. In Lau et al. [9], nasal packing controlled epistaxis in 95% (19/20) of patients. Eleven patients underwent packing with inflatable packs and 9 with nasal tampons. The individual who experienced immediate treatment failure had been packed with a nasal tampon and epistaxis was controlled after reinsertion of a second nasal pack. In the FloSeal group, epistaxis was controlled in 17 of 20 patients (85%). Of the 3 patients who experienced immediate treatment failure, inflatable nasal packs controlled bleeding in 2. When inflatable nasal packing also failed to control the remaining person's epistaxis, she underwent endoscopic sphenopalatine artery ligation. There were two readmissions within 7 days for both groups. The readmission rate was therefore 10% for both groups. Taking into account the readmission rate, the overall treatment success rate for nasal packing was 17 of 20 patients (85%); it was 15 of 20 patients (75%) for FloSeal. There was no significant difference between the primary outcomes for both groups (p = 0.73). Conversely, Khan et al. [13] evaluated FloSeal in 36 of 101 patients admitted for anterior and posterior epistaxis. The total success rate was 14% (5 of 36 cases). It was successful in 2 of 3 cases of anterior epistaxis (66%) and in only 3

of 33 cases of posterior epistaxis (9%). Fisher's exact test indicated a significant difference in the success rates of FloSeal and nasal packing in the management of posterior epistaxis (9% vs 92%, respectively; $p < 0.001$).

Tranexamic acid

Zahed et al. [19] conducted a trial to assess the efficacy of topical tranexamic compared to nasal packing. This randomised clinical trial study was conducted on 216 subjects (124 men and 92 women). Within 10 minutes of treatment, bleedings were arrested in 76 (71%) of 107 patients in the tranexamic acid group, compared with 34 (31.2%) of 109 patients in the anterior nasal packing group (odds ratio, 2.27; 95% CI, 1.68-3.06; $p < 0.001$). In addition, 102 (95.3%) of 107 patients in the tranexamic acid group were discharged in 2 hours or less vs 7 (6.4%) of 109 patients in the anterior nasal packing group. Re-bleeding was reported in 5 (4.7%) of 107 and 14 (12.8%) of 109 patients during the first 24 hours in the tranexamic acid and the anterior nasal packing groups, respectively ($p = 0.034$). After 1 week, re-bleeding in the tranexamic acid and the anterior nasal packing groups were 2.8% and 11%, respectively ($p = 0.018$). Satisfaction rate was higher with tranexamic acid (8.5 ± 1.7) than with anterior nasal packing (4.4 ± 1.8) ($p < 0.001$).

Silver nitrate

Chemical cautery using the silver nitrate has also been investigated [20 22]. Chemical cautery was the most commonly used treatment modality for anterior epistaxis (77.1%) in a cohort of 147 individuals [20], and non-dissolvable packing was the most common intervention for posterior epistaxis (78.6%). The failure rate was 23.8% for all patients undergoing chemical cautery. Among patients with anterior epistaxis who underwent chemical cautery, the failure rate was 21.0%. All 3 patients who received chemical cautery for posterior epistaxis (posterior epistaxis cautery in the clinic is done with the use of an endoscope) all experienced treatment failure. The multivariate OR of failure was significantly higher for non-dissolvable packing than for chemical cautery (6.08; 95% CI, 2.17-17.09) in cases of anterior epistaxis. Of note, 4 patients underwent operative sphenopalatine ligation for their initial epistaxis episode. All of those patients had posterior bleeding that the surgeon believed was of high enough severity that it required a surgical intervention. Furthermore, Mudunuri and Murthy [22] conducted a prospective study on 114 patients. Half of patients ($n = 57$) were managed using conservative treatment. The other half ($n = 57$) were treated with silver nitrate cautery of the bleeding points/prominent vessels. In the group treated conservatively, 30% (13 cases) had at least one episode of epistaxis. In the group treated with cautery, 26% (14 cases) had at least one episode of epistaxis. No statistical comparison was done.

Endoscopic surgical procedures

Butrymowicz et al. [12] proposed an innovative surgical treatment to control recalcitrant anterior epistaxis based on anatomical dissections, computed tomography (CT) scan and small cohort of patients. The authors evaluated the feasibility and effectiveness of surgical ligation of the greater palatine artery endoscopically. They treated surgically, after careful cadaveric dissections and CT scan analyses, 4 patients with reasonable efficacy and feasibility of the procedure.

Shrestha [14] described his personal results on 12 patients with epistaxis managed with endoscopical sphenopalatine artery ligation under general anaesthesia. Three of 12 patients developed synechia. However, the success rate in controlling epistaxis was 100%.

George et al. [23] investigated the efficacy sphenopalatine, anterior ethmoid and internal maxillary artery intervention in 25 patients. This series included 19 primary endoscopic sphenopalatine artery ligations, and three cases required adjuvant anterior ethmoid artery surgery. Exclusive anterior ethmoid artery ligation was done in two subjects and was indicated when a suspicious anterior bleed refractory to packing or cautery was present. One patient underwent embolisation of the internal maxillary artery because of high anaesthetic risk. Patients waited on average 1.9 days for the operating theatre whilst undergoing conservative management for epistaxis. The success rate for primary endoscopic sphenopalatine artery ligation surgery on the same admission was 89.4% (17/19). 80% (20/25) of patients had no further epistaxis episodes following their primary surgery, and so after discharge the overall success rate was 88% (22/25).

In a prospective study, Gandomi et al. [16] endoscopically coagulated the sphenopalatine artery in 27 patients. Three patients required bilateral cauterisation. All operations involved cauterisation with bipolar diathermy. No patient suffered recurrent epistaxis within the first 24 hours of surgery (immediate post-op. period). Three patients suffered recurrent epistaxis within 2 weeks following surgery (early post-op. period), two of whom needed anterior nasal packing with the third requiring no medical attention. One further patient had recurrent epistaxis within 7 days (early post-op. period) and also 2 months after surgery (late post-op. period). This patient further responded to conservative management including local cautery and anterior nasal packing. None of these four patients required any new surgical intervention. Therefore, the success rate was 87%. Furthermore, in another prospective study, Eladl et al. [28] showed a success

rate of 100% and no recurrence of epistaxis in a cohort of 42 patients after endoscopic sphenopalatine cauterisation. Post-operative subjective evaluation of eye dryness indicated no dryness in 90.5% of patients (n = 38) and mild dryness in only 9.5% (four patients, who complained of mild eye burning and congestion). Objective evaluation of eye dryness (using Schirmer's test) was negative in all cases. Post-operative subjective evaluation of nasal dryness indicated that dryness was present in 81% of patients (n = 34), but was severe in only 9.5% (n = 4), and resolved with medical treatment (i.e. nasal debridement and irrigation). Post-operative subjective evaluation for nasal sensation (i.e. para-aesthesia or hypoaesthesia of the nose, upper teeth and palate) indicated numbness and para-aesthesia of the upper teeth in three patients. Objective evaluation of nasal sensation found hypoaesthesia of the nasal mucosa in eight patients, but without any patient complaint. No hypoaesthesia of the hard palate was detected. No major post-operative complications or irreversible damage was encountered. Similar results, achieving the control rate of 93%, were described in the study of Minni et al. [31] (42 sphenopalatine artery and 6 anterior ethmoidal artery ligations).

Endovascular embolisation

Gottumukkala et al. [18] reviewed their experience with routine multivessel embolisation for intractable idiopathic epistaxis in 84 patients and examined the association between the number of vessels receiving embolisation and treatment outcomes. Embolisation of bilateral internal maxillary artery (IMA) with or without facial artery embolisation was performed in 76 patients (41 with one or both facial arteries, 35 without facial artery). In cases where embolisation of three vessels was performed, the facial artery treated was always ipsilateral to the bleeding site. In 10 patients, it was unclear which side was the source of bleeding. Four of these patients received bilateral IMA and bilateral facial artery embolisation, whereas six received bilateral IMA embolisation alone. Successful treatment (no re-bleeding within 30 days) was achieved in 75 patients (89%). Of the nine patients who had early re-bleeding, 6 experienced re-bleeding within 1 day of embolisation and did not require additional hospital admission. All nine patients required some form of retreatment, including surgical ligation of the ipsilateral anterior or posterior ethmoidal arteries (n = 6), endoscopic electrocautery (n = 4), and repeat embolisation (n = 1). There was one major complication (1%). A patient who underwent bilateral IMA and bilateral facial artery embolisation experienced skin sloughing of the chin, submandibular pain and oedema that caused difficulty swallowing, and severe lip oedema with mild ulceration. The oedema, pain and ulceration subsided by 1-month follow-up; the skin sloughing resolved by 8 weeks. Minor complications occurred in 22 patients (26%).

In another retrospective study [25], 16 patients were evaluated. After embolisation, immediate cessation of bleed was achieved in all 16 (100%) patients. However, in 2 (12.5%) of cases angioembolisation had to be repeated; in 1 patient on the same day and in another patient after a month. Thus, the overall success rate was 87.5%. In majority of cases (56.25%), ipsilateral IMA was the bleeding source and was embolised.

Lesley et al. [27] studied the efficacy and safety profile of detachable platinum fibered coil embolisation in a cohort of 20 patients. Seventeen patients underwent bilateral IMA embolisation. Three patients had unilateral IMA embosurgery of which two had previously undergone contralateral IMA open ligation for remote, but severe bouts of epistaxis. During the 30-day follow-up period, 95% of the cohort remained free of recurrent nose bleeding that required medical or surgical intervention. Open surgical anterior ethmoid cauterisation was needed to control bleeding in one patient (5%) who re-bled 4 days after embosurgery. 95% of patients had no complications, while one patient (5%) had short-lasting, transient unilateral facial pain.

Strach et al. [29] evaluated 48 patients suffering from intractable epistaxis underwent angiography and selective intra-arterial intervention. In this series, a total of 31 individuals presented with idiopathic epistaxis. The success rate after first-time embolisation was 29 of 31 (93.5%). After ineffective unilateral embolisation of the sphenopalatine artery in one patient (1/31; 3.2%), second-phase embolisation of the contralateral sphenopalatine, the descending palatine and branches of the facial artery, and, in a third phase, the ipsilateral sphenopalatine artery was performed, which finally lead to cessation of epistaxis increasing the success rate in group 1 to 96.8% (30/31). Despite bilateral embolisation of the sphenopalatine arteries, one patient (1/31; 3.2%) had to undergo surgical coagulation of the ethmoidal artery to achieve haemostasis. One patient with initially successful unilateral embolisation of the sphenopalatine artery experienced reoccurrence of epistaxis after an asymptomatic period of 14 days and underwent additional septoplasty, thereby achieving haemostasis. Thus, the overall long-term success rate was 93.5%.

Laser

Zhang and Qiu [26] conducted a prospective, randomised, single-blinded study on the use of Nd:YAG laser for treatment of anterior epistaxis. A total of 80 consecutive patients were included and equally divided into Laser and ointment groups. After 12 weeks, 85% of laser patients

and 40% of control patients had no reported bleeding. The outcome score of the 80 patients at 4 weeks after treatment showed no significant differences between groups (mean rank: 43.93 vs 37.08, ointment group versus laser group, $p = 0.130$). However, the outcome score at 12 weeks after treatment showed a significant difference between the two groups (mean rank: 49.60 vs 31.40, ointment group versus laser group, $p < 0.01$). Neither experimental group had complications, such as blood transfusion, hospitalisation, visible nasal scars, nasal adhesions, or nasal septum perforation.

Discussion

The purpose of this study was to examine recent concepts in managing idiopathic epistaxis. Nasal packing still represents the first-line approach to epistaxis, although, at present, it appears that there is clear evidence in the literature suggesting that it is less effective and associated with more admissions and longer hospital stays than endoscopic electrocoagulation-based management of epistaxis. In 65% to 70% of cases of epistaxis, simple first aid measures provided by the primary care physician or emergency physician are effective. If bleeding persists, patients should be urgently referred to the ENT Department. So long as the source of the bleeding is visible, most cases of epistaxis can be successfully treated using electrical or chemical cautery. For posterior epistaxis, surgical intervention is markedly superior to packing. The method of choice is endoscopic clipping or coagulation of the sphenopalatine artery, which controls bleeding in 98% of cases [32]. Early sphenopalatine ligation has a cost-effective profile, while a randomised prospective trial of early ligation vs nasal packing found an overall cost-savings of approximately $ 7080. Additionally, the length of stay was less than half that for nasal packing. A 2003 retrospective study by Klotz et al. [33] found that ligation had a mean length of stay of 2.1 days and $ 3851 of associated costs. Moreover, Henderson et al. [15] conducted an analysis on costs before and after application of a protocol teaching junior physicians on the use of cauterisation instead of nasal packing. They found a per-patient average saving of £ 117, and per year £ 43,345 assuming an average of 31 presentations per month. Its cost-effectiveness was also confirmed in the management of posterior epistaxis with endoscopic sphenopalatine ligation instead of traditional nasal packing. Dedhia et al. [34] found, in the base case, costs for first-line endoscopic sphenopalatine ligation and current practice (nasal packing) arms were $ 6450 and $ 8246, respectively. In other words, there was a cost savings of $ 1796 in the first-line endoscopic sphenopalatine ligation arm.

On the other hand, endovascular interventions are an important option for the treatment of life-threatening posterior epistaxis. Percutaneous embolisation is highly efficient and its safety profile compares favourably with alternative therapeutic modalities. It is noteworthy that, unlike other treatment strategies, major complications, such as cerebrovascular embolism or soft-tissue damage, may occur. Hence, this approach should be the last option and to be used only when all other options fail or in the presence of severe comorbidities that might preclude general anaesthesia [35].

Finally, data concerning the use of drugs (i.e. tranexamic acid) or other medical devices did not show a substantially stronger clinical applicability. This is due to the fact that the data are extrapolated from few studies usually with a small cohort of patients, and most are neither randomised nor comparative.

Conclusions

Currently, the most common first-line treatment of idiopathic epistaxis is nasal packing, although there is a clear trend away from the use of nasal packs. Although it is a quick and easy to learn technique, emerging evidence show that cauterisation provides economic advantages and is easy to teach, especially for anterior epistaxis, to non-otolaryngologists. Early sphenopalatine artery ligation should also confer financial advantages, while the use of endovascular techniques should be reserved as a last option. Further research is urgently needed to assess the efficacy of new biomaterials that might have a significant impact on first-line management.

Conflict of interest statement

None declared.

References

1 Rockey JG, Anand R. *A critical audit of the surgical management of intractable epistaxis using sphenopalatine artery ligation/diathermy.* Rhinology 2002;40:147-9.

2 Al Khtoum N, Al Roosan M. *The evaluation of conservative measures in the treatment of epistaxis.* Khartoum Med J 2008;1:15-7.

3 Bray D, Giddings CE, Monnery P, et al. *Epistaxis: are temperature and seasonal variations true factors in incidence?* J Laryngol Otol 2005;119:724-6. https://doi.org/10.1258/0022215054798032.

4 Comelli I, Vincenti V, Benatti M, et al. *Influence of air temperature variations on incidence of epistaxis.* Am J Rhinol Allergy 2015;29:175-81. https://doi.org/10.2500/ajra.2015.29.4176.

5 Purkey MR, Seeskin Z, Chandra R. *Seasonal variation and predictors of epistaxis.* Laryngoscope 2014;124:2028-33. https://doi.org/10.1002/lary.24679.

6 Bertrand B, Eloy P, Rombaux P, et al. *Guidelines to the management of epistaxis.* B-ENT 2005;Suppl 1:27-41.

7 Ho EC, Chan JY. *Front-line epistaxis management: let's not forget the basics.* J Laryngol Otol 2008;122:696-9. https://doi.org/10.1017/S0022215108002181.

8 Whiting PF, Rutjes AW, Westwood ME, et al. *QUADAS-2: a revised tool for the quality assessment of diagnostic accuracy studies.* Ann Intern Med 2011;155:529-36. https://doi.org/10.7326/0003-4819-155-8-201110180-00009.

9 Lau AS, Upile NS, Lazarova L, et al. *Evaluating the use of Floseal haemostatic matrix in the treatment of epistaxis: a prospective, control-matched longitudinal study.* Eur Arch Otorhinolaryngol 2016;273:2579-84. https://doi.org/10.1007/s00405-016-3948-y.

10 Zou Y, Deng YQ, Xiao CW, et al. *Comparison of outcomes between endoscopic surgery and conventional nasal packing for epistaxis in the posterior fornix of the inferior nasal meatus.* Pak J Med Sci 2015;31:1361-5. https://doi.org/10.12669/pjms.316.8340.

11 Iimura J, Hatano A, Ando Y, et al. *Study of hemostasis procedures for posterior epistaxis.* Auris Nasus Larynx 2016;43:298-303. https://doi.org/10.1016/j.anl.2015.09.015.

12 Butrymowicz A, Weisstuch A, Zhao A, et al. *Endoscopic endonasal greater palatine artery cauterization at the incisive foramen for control of anterior epistaxis.* Laryngoscope 2016;126:1033-8. https://doi.org/10.1002/lary.25677.

13 Khan MK, Reda El Badawey M, Powell J, et al. *The utility of FloSeal haemostatic agent in the management of epistaxis.* J Laryngol Otol 2015;129:353-7. https://doi.org/10.1017/S0022215115000663.

14 Shrestha BL. *Endoscopic sphenopalatine artery cauterization in recurrent posterior epistaxis: an experience at Dhulikhel Hospital, Kathmandu University Hospital.* Kathmandu Univ Med J (KUMJ) 2014;12:85-6.

15 Henderson AH, Larkins A, Repanos C. *The use of bipolar electrocautery in adult epistaxis management: using audit of one hundred and twenty-four cases to define a standardised protocol.* Clin Otolaryngol 2013;38:554-8. https://doi.org/10.1111/coa.12191.

16 Gandomi B, Arzaghi MH, Khademi B, et al. *Endoscopic cauterization of the sphenopalatine artery to control severe and recurrent posterior epistaxis.* Iran J Otorhinolaryngol 2013;25:147-54.

17 Dutta S, Mukherjee A, Saha J, et al. *Modified technique of anterior nasal packing: a comparative study report.* Indian J Otolaryngol Head Neck Surg 2012;64:341-5. https://doi.org/10.1007/s12070-011-0343-2.

18 Gottumukkala R, Kadkhodayan Y, Moran CJ, et al. *Impact of vessel choice on outcomes of polyvinyl alcohol embolization for intractable idiopathic epistaxis.* J Vasc Interv Radiol 2013;24:234-9. https://doi.org/10.1016/j.jvir.2012.10.001.

19 Zahed R, Moharamzadeh P, Alizadeharasi S, et al. *A new and rapid method for epistaxis treatment using injectable form of tranexamic acid topically: a randomized controlled trial.* Am J Emerg Med 2013;31:1389-92. https://doi.org/10.1016/j.ajem.2013.06.043.

20 Shargorodsky J, Bleier BS, Holbrook EH, et al. *Outcomes analysis in epistaxis management: development of a therapeutic algorithm.* Otolaryngol Head Neck Surg 2013;149:390-8. https://doi.org/10.1177/0194599813492949.

21 Kilty SJ, Al-Hajry M, Al-Mutairi D, et al. *Prospective clinical trial of gelatin-thrombin matrix as first line treatment of posterior epistaxis.* Laryngoscope 2014;124:38-42. https://doi.org/10.1002/lary.24240.

22 Mudunuri RK, Murthy MA. *The treatment of spontaneous epistaxis: conservative vs cautery.* J Clin Diagn Res 2012;6:1523-4. https://doi.org/10.7860/JCDR/2012/4215.2548.

23 George A, Smatanova K, Joshi H, et al. *Sphenopalatine, anterior ethmoid and internal maxillary artery intervention in the management of refractory epistaxis: their efficacy in 25 patients.* Clin Otolaryngol 2012;37:321-5. https://doi.org/10.1111/j.1749-4486.2012.02499.x.

24 Kourelis K, Shikani AH. *Effectiveness of chitosan-based packing in 35 patients with recalcitrant epistaxis in the context of coagulopathy.* Clin Otolaryngol 2012;37:309-13. https://doi.org/10.1111/j.1749-4486.2012.02488.x.

25 Baloch MA, Awan MS, Resident HN. *Angioembolization in intractable epistaxis - a tertiary care experience.* J Pak Med Assoc 2012;62:254-6.

26 Zhang J, Qiu R. *A prospective randomised controlled trial of Nd:YAG laser photocoagulation versus liquid paraffin plus antiseptic cream in the treatment of recurrent epistaxis.* Clin Otolaryngol 2012;37:271-5. https://doi.org/10.1111/j.1749-4486.2012.02509.x.

27 Lesley WS, Rangaswamy R, Patel DV. *Results of epistaxis embosurgery using detachable platinum fibered coils.* J Neurointerv Surg 2010;2:171-5. https://doi.org/10.1136/jnis.2009.001834.

28 Eladl HM, Elmorsy SM, Khafagy YW. *Endoscopic devascularisation of sphenopalatine bundle in intractable posterior epistaxis: technique, efficacy and safety.* J Laryngol Otol 2011;125:1136-40. https://doi.org/10.1017/S0022215111002180.

29 Strach K, Schröck A, Wilhelm K, et al. *Endovascular treatment of epistaxis: indications, management, and outcome.* Cardiovasc Intervent Radiol 2011;34:1190-8. https://doi.org/10.1007/s00270-011-0155-5.

30 Côté D, Barber B, Diamond C, et al. *FloSeal hemostatic matrix in persistent epistaxis: prospective clinical trial.* J Otolaryngol Head Neck Surg 2010;39:304-8.

31 Minni A, Dragonetti A, Gera R, et al. *Endoscopic management of recurrent epistaxis: the experience of two metropolitan hospitals in Italy.* Acta Otolaryngol 2010;130:1048-52. https://doi.org/10.3109/00016481003621538.

32 Beck R, Sorge M, Schneider A, et al. *Current approaches to epistaxis treatment in primary and secondary care.* Dtsch Arztebl Int 2018;115:12-22. https://doi.org/10.3238/arztebl.2018.0012.

33 Klotz DA, Winkle MR, Richmon J, et al. *Surgical management of posterior epistaxis: a changing paradigm.* Laryngoscope 2002;112:1577-82. https://doi.org/10.1097/00005537-200209000-00008.

34 Dedhia RC, Desai SS, Smith KJ, et al. *Cost-effectiveness of endoscopic sphenopalatine artery ligation versus nasal packing as first-line treatment for posterior epistaxis.* Int Forum Allergy Rhinol 2013;3:563-6. https://doi.org/10.1002/alr.21137.

35 Goddard JC, Reiter ER. *In patient management of epistaxis: outcomes and cost.* Otolaryngol Head Neck Surg 2005;132:707-12. https://doi.org/10.1016/j.otohns.2005.02.001.

Received: March 18, 2018 - Accepted: May 27, 2018

How to cite this article: Meccariello G, Georgalas C, Montevecchi F, et al. *Management of idiopathic epistaxis in adults: what's new?* Acta Otorhinolaryngol Ital 2019;39:211-219. https://doi.org/10.14639/0392-100X-2155

Address for correspondence: Giuseppe Meccariello, Department of Head-Neck Surgery, Otolaryngology, Head-Neck and Oral Surgery Unit, Morgagni Pierantoni Hospital, via Carlo Forlanini 34, 47100 Forlì, Italy. E-mail: drmeccariello@gmail.com

ACTA OTORHINOLARYNGOLOGICA ITALICA 2019;39:220-229; doi: 10.14639/0392-100X-2185

HEAD AND NECK

Differential isoform expression of SERCA and myosin heavy chain in hypopharyngeal muscles

Espressione differenziale di isoforma del SERCA e delle catene pesanti della miosina nella muscolatura ipofaringea

MOHAMMED ELRABIE AHMED[1 2], H. BANDO[1], S. FUSE[1], H. MOSTAFA ABDELFATTAH[3], MONA ELRABIE AHMED[4], M. ABDEL-KADER AHMED[2], T. TSUJIKAWA[1], Y. HISA[1 5]

[1] Department of Otolaryngology-Head and Neck Surgery, Kyoto Prefectural University of Medicine, Kyoto, Japan; [2] Department of Otorhinolaryngology-Head and Neck Surgery, Sohag University Hospitals, Faculty of Medicine, Sohag University, Sohag, Egypt; [3] Department of Otorhinolaryngology-Head and Neck Surgery, Alexandria University, Alexandria, Egypt; [4] Unit of Phoniatrics-Department of Otorhinolaryngology-Head and Neck Surgery, Sohag University, Egypt; [5] Department of Speech and Hearing Sciences and Disorders, Kyoto Gakuen University, Japan

SUMMARY

Composition of slow, fast and hybrid fibres of pharyngeal muscles, associated with pharyngeal movements and regulation, has been rarely studied. The present study aimed to identify expression of sarcoplasmic reticulum $Ca2^+$ ATPase (SERCA) and myosin heavy chain (MHC) and hybrid isoforms in different pharyngeal muscles of young and aged rats as well as humans. Isoform expression profiles of SERCA, MHC and hybrid isoforms among six components of pharyngeal muscle were immunohistochemically evaluated in rat and human. The result showed that pharyngeal muscles predominantly expressed fast fibres (SERCA1 and MHCII), whereas expression of slow fibres (SERCA2 and MHCI) was low, but different depending on muscle components. Inner layer of pharyngeal muscles expressed more SERCA2 and hybrid fibres than the outer layer. Pharyngeal muscles in aged rats showed increased hybrid fibers and SERCA2. Human thyropharyngeus also showed a higher portion of fast fibres compared to cricopharyngeus. Thus, in contrast to abundance of fast fibres, slow and hybrid fibres are differentially expressed depending on muscle components and layers as well as aging. These results lead to further understanding of coordinated regulation for speech and swallowing. The unique data presented in this study on SERCA isoform expressions in both rats and human suggest an ability to handle calcium changes according functional demands.

KEY WORDS: Hypopharynx • SERCA • Myosin heavy chain • Isoform • Aging

RIASSUNTO

La correlazione fra la funzione faringea e la composizione delle fibre lente, rapide ed ibride della muscolatura faringea è stata scarsamente studiata. Questo lavoro si propone di studiare l'espressione del SERCA, MHC ed isoforme ibride muscolari in topi giovani, di età avanzata ed anche in pazienti attraverso metodiche di immunoistochimica. I risultati evidenziano una maggior espressione nella muscolatura faringea delle fibre muscolari veloci SERCA1 e MHCII. Lo strato interno esprime maggiormente SERCA2 e fibre ibride rispetto allo strato esterno. La muscolatura faringea dei topi di età avanzata mostra un aumento di espressione delle fibre ibride e SERCA2. Le fibre muscolari veloci, lente ed ibride sono pertanto espresse in misura diversa a seconda dei fasci muscolari, strati ed età dei pazienti e potrebbero giocare un ruolo nella coordinazione del linguaggio e deglutizione. Questi dati suggeriscono inoltre una possibile variabilità nello sfruttamento del calcio a seconda delle richieste funzionali.

PAROLE CHIAVE: *Ipofaringe • SERCA • Catena pesante della miosina • Isoforma •Invecchiamento*

Introduction

Pharyngeal muscles play a crucial role in the deglutition process by applying the proper pressures required to receive and propel swallowed material (bolus) through the pharynx and upper oesophageal sphincter (UES), and to shape the airway to modulate resonance during voice and speech production [1]. In fact, the pharyngeal stage of swallowing is involuntary, and is the most rapid and complex phase in the entire deglutition process that requires bilateral sequenced activation and inhibition of more than 30 pairs of different muscles of the mouth, pharynx, larynx and oesophagus [2]. All these different muscles need to be adaptable to boluses of differing volumes, consistency and rheological characteristics.

Moreover, UES prevents reflux of oesophageal contents

into the pharynx to guard airway aspiration and prevent air from entering the oesophagus while breathing. It is generally agreed that the cricopharyngeus is a major contributor to the UES (thyropharyngeal (TP) muscle in animals), and the cranial cervical oesophagus also contributes to it in its proximal and distal extents. These muscles have unique rich innervation (1:2-1:6 nerve-to-muscle fibre innervation ratio) compared with limb and extraocular eye muscles, which is important for the fine control required for their highly specialised and complex functions [3].

Weakness and fatigue of pharyngeal muscles due to aging or any other factor may result in risks of malnutrition and/or aspiration pneumonia because of either miss-direction into the lungs or retention of pharyngeal residue that poses an increased aspiration risk [4 5]. Indeed, normal pharyngeal deglutition constitutes the most important element in a safe swallowing process.

For the above reasons, understanding the anatomical, physiological and biological characteristics of pharyngeal muscles and their functions in the deglutition process has widely attracted the attention of scientists for decades. However, much remains unknown with respect to the distributions of the different fibre types in the pharyngeal muscles as well as the different muscle functional behaviours. What can be currently inferred from the available information is that pharyngeal muscles are very different from the extensively studied limb and trunk musculatures, whose main functions are locomotion and posture, in terms of specialization that in turn produces functional differences in contraction times, tension generation, endurance and tuning of movement. A condition that permits pharyngeal muscles to engage in extremely rapid and prolonged contraction, perform highly refined contractions, constant activity even at rest, and has specific aging-related influences [6 7].

In fact, muscle fibres have very different structures, histochemistries, biochemistries and physiologies. The functional properties of each muscle are established by the speed, force, sustainability of contraction (endurance) and contraction/relaxation rate. The major proteins responsible for contraction and relaxations in skeletal muscle are sarcoplasmic reticulum $Ca2^{+}$-ATPase (SERCA) and myosin heavy chain (MHC) [8 9]. SERCA plays the main role in the muscle contraction/relaxation cycle [10], while MHC in muscle fibres regulates the contraction force and velocity. Consequently, the difference between contraction and relaxation times of fast and slow muscle fibres is linked to different SERCA and/or MHC isoform expression: fast fibres express SERCA1 and MHCII, whereas slow fibres express SERCA2 and MHCI [11]. Fast muscle fibres (type II) are recruited for short maximal efforts with fast contraction speeds and are fatigued easily. In contrast, slow muscle fibres (type I) have a slow contraction speed and are highly resistant to fatigue. Thus, muscle regions with a high proportion of fast type II fibres facilitate rapid and phasic movements, whereas those with a high proportion of type I fibres are generally involved in postural adjustments [12]. Therefore, the proportions and distribution patterns of MHC- and SERCA-containing fibres in muscle are closely related to muscle functions.

The distribution and role of SERCA and MHC isoforms in pharyngeal muscles are relatively poorly documented. Thus, the present study aimed to identify expression of SERCA, MHC and hybrid isoforms in different pharyngeal muscles of young and aged rats. SERCA isoform expression was also examined in two human pharyngeal muscles, thyropharyngeus (TP) muscle and cricopharyngeus (CP). Because of the lack of available information on the expression and role of SERCA isoforms in pharyngeal muscles, the data obtained in the current study may assist in better understanding of the physiological functions of pharyngeal muscle, age-related remodelling and their effects on pathological conditions.

Materials and methods

Animal tissue preparation

Adult male Wistar rats at 8-12 weeks of age and weighing 280-350 g were used in the current study (n = 10). Rats at 24 months of age were also employed as aged models (n = 4) (Shimizu Laboratory Supplies Co., Kyoto, Japan). Animals were anaesthetised by intraperitoneal injection of sodium pentobarbital (30-60 mg/kg) and then fixed with 4% paraformaldehyde following cardiac perfusion. The pharyngolarynxes were excised immediately and immersed in the same fixative for approximately 12 hours at 4°C. Paraffin processing of tissues was performed before preparation for microtome sectioning at 8-10 μm thicknesses.

Surgical microscopic examination and haematoxylin and eosin staining

The anatomy of rat pharyngeal muscles was studied carefully under an operating microscope (ICOM 300) by microdissection. Next, a complete set of serial transverse sections (5-8 μm thick) were obtained and mounted on glass slides. Some of these sets were stained with haematoxylin and eosin for initial close anatomical inspection of the rat pharyngeal muscles.

Immunohistochemistry

Deparaffinised sections were incubated with 3% H_2O_2 in

PBS for 10 minutes. After microwave treatment (5 minutes × 3 times at 500 W in citrate buffer, pH 6), the sections were incubated in blocking solution (0.3 M glycine, 50 mM ammonium chloride, and 1% bovine serum albumin in PBS) for 30 minutes before incubation with primary antibodies. The sections were then incubated with anti-SERCA1 or 2 and anti MHCI or II (Table I) antibodies for 12 days at 4°C, washed, and incubated with Alexa Fluor® 488 donkey anti-mouse IgG and Alexa Fluor® 594 donkey anti-goat IgG secondary antibodies for 1 hour at room temperature. Double immunohistochemistry was performed using anti-SERCA1 + anti-SERCA2, and anti-SERCA2 + anti-MHCII antibody combinations for dual detection. Negative controls (no application of the primary antibody) were used to document the extent of non-specific binding of the secondary antibody.

At the beginning of our research, we tried to perform a double IHC for MHC and SERCA study. For, MHC isoforms in frozen section (primary AB I "BA-D5"; IIa "SC-71"; IIb "BF-F3" and IIX "6H1" with secondary AB Alexa 488 IgG2b; Alexa 647 IgG1; Alexa 488 IgM; Alexa 647 IgG1 and Alexa 555 IgM respectively) but, we failed to get immunoreaction at several concentrations. Hence, we shifted to paraffin sections, the current antibodies and protocol. We tried triple IHC and experienced some difficulties for autofluorescence and poor identification of low amounts of expression of MHCI.

Immunostained tissue sections were examined using a FV-1000 laser confocal microscope (Olympus, Tokyo, Japan). ImageJ 1.46 software was used to manually count and calculate the ratio of cells positive for each antibody.

Human samples

Human TP and CP muscle specimens were used in the current study. Specimens were intra-operatively obtained from non-malignant laryngeal tissue of patients undergoing total laryngectomy for malignancy if the sampling site was not affected by malignancy or preoperative radiotherapy and specimen sampling did not interfere with diagnosis.

The study involved five male patients with a mean age of 52.7 years. All tissue samples were fixed immediately after surgery in 4% paraformaldehyde and then embedded in paraffin. Paraffin processing of all specimens and microtome sectioning at 8-10 µm was performed. Similar to rats, standard immunofluorescence staining procedures were applied to evaluate SERCA1, SERCA2 and dual expression in human TP and CP muscles. Immunostained tissue sections were examined using a FV-1000 laser confocal microscope, followed by quantification of positive cells for each antibody by ImageJ 1.46 software.

Statistical analysis

Data of the positive cell ratio for each antibody in rats and humans were statistically analysed using SPSS statistical software version 16. The unpaired Student's t-test was applied to compare differences between SERCA, MHC, and hybrid isoforms in various pharyngeal muscles of young and aged rats as well as humans. Values are presented as means ± standard deviation (SD). For all statistical tests, $p < 0.05$ was considered as significant.

Results

Anatomy of rat pharyngeal muscles

The anatomy of rat pharyngeal muscles was microscopically studied by microdissection (see Fig. 1). As reported by Kobler et al. [13], rigorous examination of pharyngeal muscle anatomy confirmed that the HP and TP muscles formed the main bulk of the rat posterior pharyngeal wall, which originated from the hyoid bone and thyroid cartilage, respectively, while both muscles inserted obliquely into the midline pharyngeal raphe. HP muscle is not defined in human and higher vertebrate animals and appears anatomically homologous to the superior and middle constrictor muscle. In addition, at the pharyngo-oesophageal junction, we observed muscle fibres that arched across the posterior pharyngeal wall to the opposite side without inserting into the midline raphe. These fibres were overlapped by the more oblique fibres of the TP muscle. According to the previous report, this muscle should be labelled as semicircular SC muscle that does not have an insertion on the cricoid cartilage through serial sections. Thus, it appeared that the rat did not have a strictly defined CP muscle, although the SC resembled it in terms of the general location and arrangement.

Table I. Antibodies used in this study.

Isoform	Antibody	Description
SERCA1	VE121G9; ab2819	Mouse monoclonal to SERCA1 IgG1 (abcam)
SERCA2	SERCA 2 (C-20):sc-8094	Goat polyclonal against peptide mapping at C-terminus of SERCA 2 (Santa Cruz Biotechnology)
MHCI	NOQ7.5.4D	Mouse monoclonal to slow myosin skeletal muscle IgG1 (abcam)
MHCII	MY-32; ab51263	Mouse monoclonal to fast myosin skeletal muscle IgG1(abcam)

Immunolocalisation of myosin and SERCA isoforms in young rat pharyngeal muscle

Expression of myosin (MHCI and II) and SERCA (1 and 2) isoforms and hybrid isoforms were evaluated in the six selected pharyngeal muscle components in young rats. The studied hypopharyngeal muscle components were hyopharyngus (HP), thyropharyngeus (TP), semicircular (SC) muscles, palatopharyngeus (PP), soft palate and cervical oesophagus (CE). SERCA1 and MHCII, which are considered as fast fibres, were predominant and similarly expressed in all hypopharyngeal muscles, compared with slow muscle fibres (MHCI or SERCA2) (Fig. 2, Table II). Expression of slow fibres was basically low in all examined muscles, but slow fibres showed differential patterns depending on muscle components, where SERCA2 expression was found in all muscles in high proportion than the corresponding slow MHCI which was observed only in HP, TP and SC (Table II). Hybrid fibres (SERCA1 + 2 or MHCII + SERCA2) were detected in all pharyngeal muscles which may assist their complex functional demands. SERCA/MHC mismatches (mainly SERCA2/MHCI; HP, 12.9 ± 1.7/6.2 ± 1; TP, 14.3 ± 2.3/5.5 ± 1.6; SC, 30.9 ± 2.7/9.2 ± 1.4) were found in normal rats, which usually occur in some pathological muscle diseases such as muscle disuse and denervation. This suggests that calcium handling fibre types are regulated differently than that for myosin (Table II). Furthermore, we evaluated potential differences of SERCA isoforms in muscular layers, comparing inner submucosal layer versus outer layer in SC and CE muscles. The inner layer expressed more SERCA2 and hybrid fibres than the outer layer, although sharp delineation between the two layers was indistinct (Fig. 3). Those results indicate that the vast majority of pharyngeal muscles were regulated by fast fibres, together with differentially coordinated distribution of slow and hybrid fibres in pharyngeal muscle components as well as muscular layers, which may assist their complex functional demands in pharyngeal kinetics.

Aging-related changes in pharyngeal muscles

To understand potential effects of aging on pharyngeal muscles, we performed comparative analysis of MHC and SERCA, and hybrid isoforms in young and aged rats. We found that aged rats showed higher ratios of hybrid fibres in all pharyngeal muscle components than young rats (Table II). Among slow fibres, SERCA2 was increased during aging, whereas MHCI was decreased (Fig. 4 and Table II). These observations suggest the presence of aging-related changes in pharyngeal muscles, where an increase of hybrid fibres and differential slow fibre isoforms was observed.

SERCA expression in human hypopharyngeal muscles

Finally, we verified our findings in rats by evaluating human tissues derived from clinical specimens by immunohistochemistry. SERCA isoforms were analysed in CP muscles, in consideration of anatomical differences between human and rat since rats possess SC, but not a counterpart corresponding to CP in humans. SERCA1

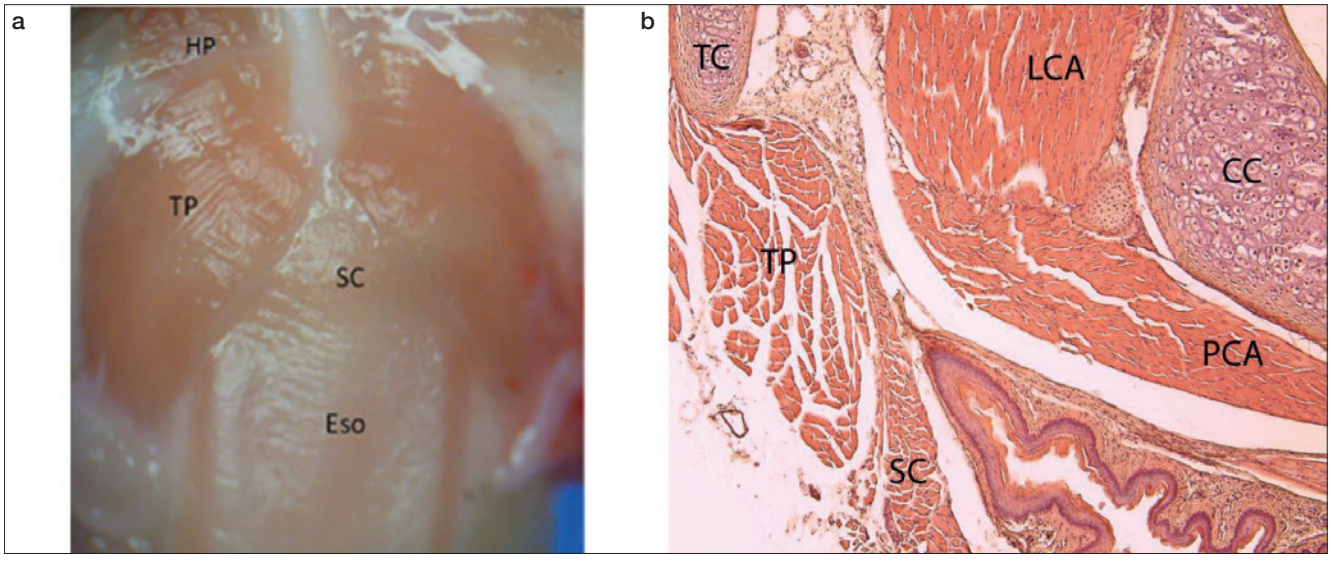

Fig. 1. Anatomical findings of rat pharyngeal muscles. (**a**) Posterior view of the pharynx showing hyopharyngus (HP), thyropharyngeal (TP) and semicircular (SC) muscles, and the cervical oesophagus (Eos). (**b**) Haematoxylin and eosin staining (× 200) showing SC muscle attachment to thyroid cartilage (TC), LCA (lateral cricoarytenoid), and PCA (posterior cricoarytenoid).

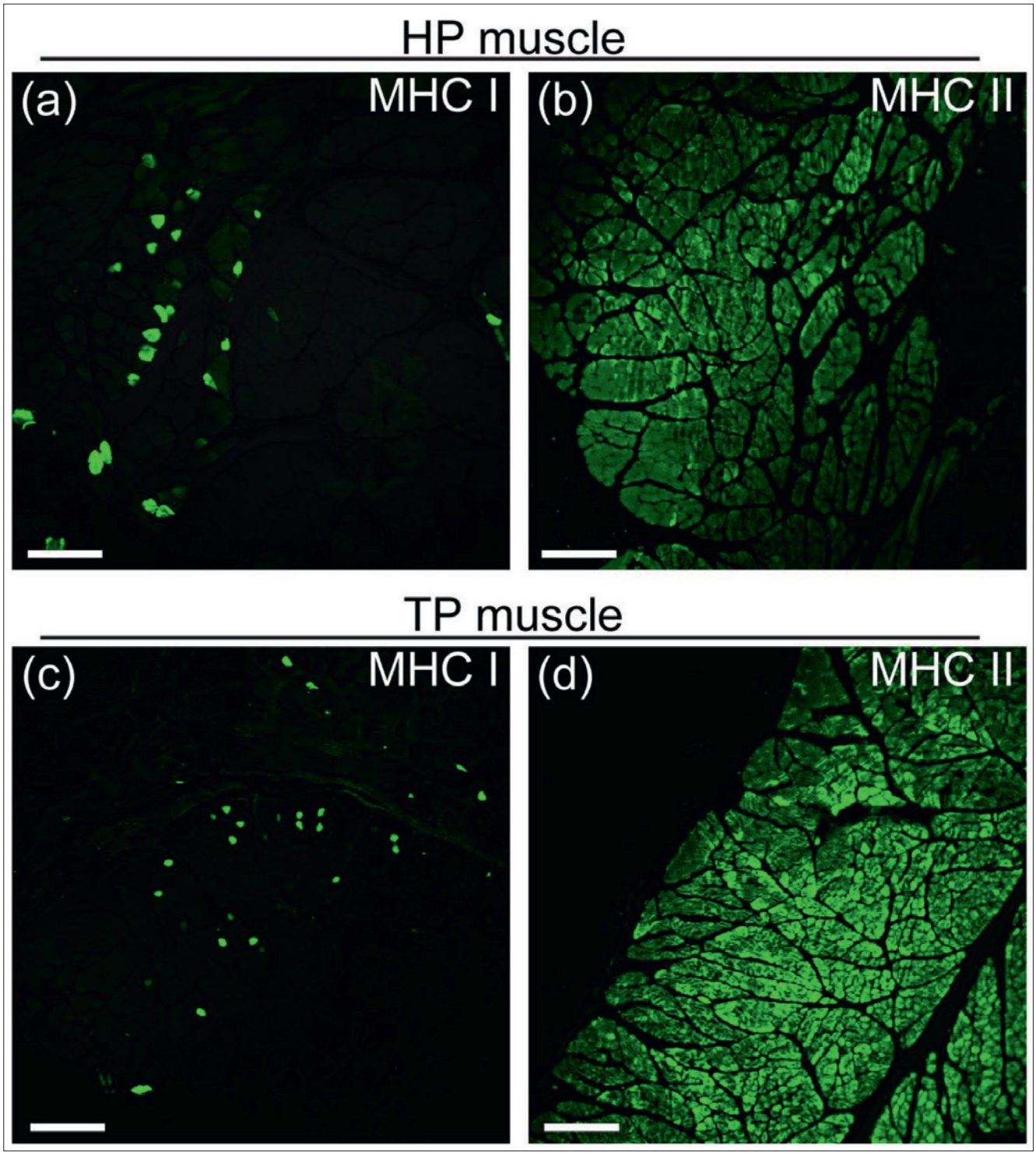

Fig. 2. Myosin heavy chain (MHC) isoform expression in hyopharyngus (HP) and thyropharyngeal (TP) muscles. (**a**) MHCI expression (slow fibres) in HP muscle, (**b**) MHCII (fast fibres) in HP muscle, (**c**) MHCI expression (slow fibres) in TP muscle, and (**d**) MHCII (fast fibres) in TP muscle; young rats, ×100. Scale bar = 50 μm.

was less predominant in human CP muscle in comparison with human TP muscle (74 ± 10.1 in TP vs 47.7 ± 14.5 in CP). However, human CP muscle contained more SERCA2 (61.9 ± 14.9) and hybrid fibres (15.6 ± 3.3) than human TP muscle (Fig. 5a and Table III). Next, we analysed isoform expression in TP muscles in which humans

Table II. Summary of the ratios of cells positive (mean ± SD) for SERCA1, SERCA2, MHCI, MHCII and hybrids in young and aged rat hypopharyngeal muscles and the results of unpaired t-tests.

	SERCA1	SERCA2	MHCI	MHCII	Hybrid (S1+S2)	Hybrid (MII+S2)
Hyopharyngus						
Young (mean ± SD)	94 ± 0.4	12.9 ± 1.7	6.2 ± 1	94.5 ± 1	10.4 ± 4	11.5 ± 2.9
Aged (mean ± SD)	97.9 ± 0.7	17.8 ± 2.2	4.8 ± 1.7	97.5 ± 0.6	16.7 ± 0.8	18.3 ± 0.6
Young vs aged (P value)	< 0.05	< 0.05	NS	< 0.05	< 0.05	< 0.05
Thyropharyngus						
Young (mean ± SD)	95.4 ± 1	14.3 ± 2.3	5.5 ± 1.6	94.9 ± 1.2	12 ± 2.7	12 ± 2.2
Aged (mean ± SD)	97.9 ± 1	16.9 ± 1.5	3.6 ± 0.3	98.1 ± 1	14 ± 2	14.3 ± 2.4
Young vs aged (P value)	< 0.05	< 0.05	<0.05	< 0.05	< 0.05	< 0.05
Semicircular muscle						
Young (mean ± SD)	90.7 ± 1.4	30.9 ± 2.7	9.2 ± 1.4	91 ± 1.4	19.7 ± 2.7	17.4 ± 2.8
Aged (mean ± SD)	92.7 ± 1.9	34.5± 2.5	6.6 ± 1.5	94.5 ± 0.9	25.4 ± 1	26.3 ± 6
Young vs aged (P value)	NS	< 0.05	< 0.05	< 0.05	< 0.05	< 0.05
Cervical oesophagus						
Young (mean ± SD)	100	26.1 ± 2.5	0	100	—	—
Aged (mean ± SD)	100	30.8 ± 2.5	0	100	—	—
Young vs aged (P value)	NS	< 0.05	NS	NS	—	—
Palatopharyngeus						
Young (mean ± SD)	100	19.2 ± 3.5	0	100	18 ± 4.2	18.3 ± 4.5
Aged (mean ± SD)	99 ± 0.8	28.4 ± 2.4	1 ± 0.9	99.3 ± 0.7	28.4 ± 4.2	28.5 ± 3
Young vs aged (P value)	NS	< 0.05	NS	NS	< 0.05	< 0.05
Soft palate						
Young (Mean ± SD)	100	15.7 ± 2.3	0	100	15.6 ± 3	15 ± 2
Aged (mean ± sD)	99.7 ± 0.5	19.1 ± 2.2	0.9 ± 0.8	99.6 ± 0.5	19 ± 1.7	18.8 ± 2.7
Young vs aged (P value)	NS	NS	NS	NS	NA	NS

P < 0.05 was considered as statistically significant; SD: standard deviation.

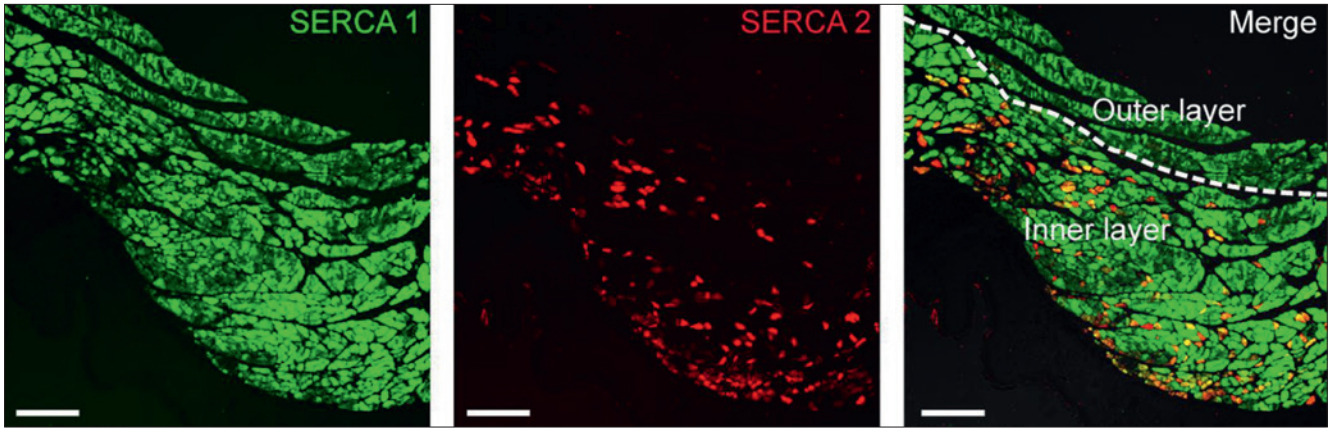

Fig. 3. Differential expression of SERCA isoforms in inner and outer layers of semicircular muscle. SERCA1 (green) was more apparent in the outer layer, while SERCA2 (red) was apparent in the inner layer. Scale bar = 50 μm.

and rats have anatomical analogy. Human TP muscles showed higher expression of SERCA1 than SERCA2. Those observations were similar to our findings in rats, although SERCA2 was relatively high in human TP muscles (74 ± 10.1 vs 37.5 ± 11.8) (Fig. 5b , Table III), which also leads to the notion that mean age in human samples

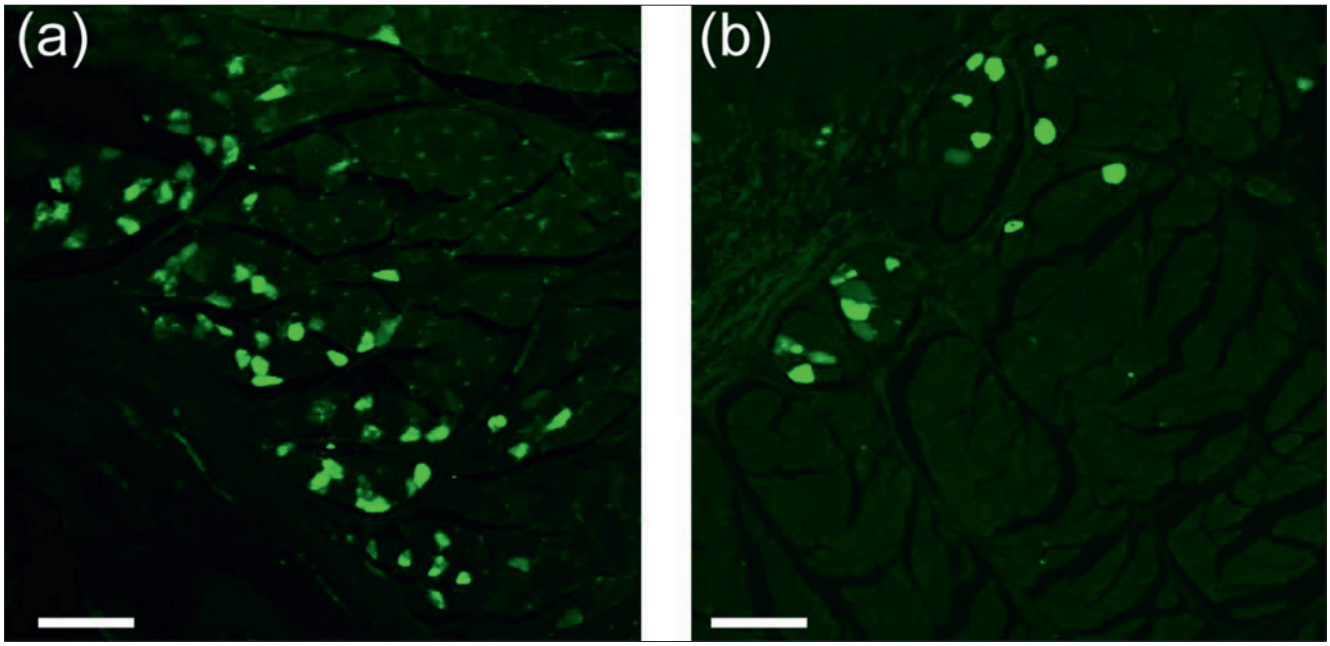

Fig. 4. Age-related decrease of MHCI in SC muscle: (**a**) MHCI in young rats at 8-12 weeks, and (**b**) MHCI in aged rats at 24 months. Expression of MHCI in aged mice decreased in SC muscle. Scale bar = 50 μm.

is 52.7 years. These data support our observations in rats and highlight differential isoform expression depending on the components of hypopharyngeal muscles.

Discussion

By expression analysis of SERCA and MHC isoforms in multiple components of pharyngeal muscles, this study has provided further insights into understanding fibre types and their particular functions, which are potentially associated with muscle types and aging. As shown in Table II, all examined pharyngeal muscles predominantly contained fast fibres (SERCA1 and MHCII) at almost similar high ratios (90-100%), while showing a lower proportion (0-35%) of SERCA2 and MHCI (slow fibre) expression. SERCA2 was observed at a significantly lower proportion in all muscles, while MHCI was localised only in pharyngeal constrictor (HP and TP) and SC muscles, and not detected in other muscles. However, the rat CE striated muscle was found to contain only fast twitch fibres (MHCII and SERCA1), which is inconsistent with previous observations [14]. In fact, to the best of our knowledge, the present SERCA isoform data represent a unique data set that supplements the few relevant data.

The composition of MHC isoforms in pharyngeal muscle is still controversial. Our results of MHCI isoform profiles supported early data in the literature. Bonington et al. [15] reported that MHCI accounted for approximately 10% of the SC muscle, which is similar to the ratio found in the current study. In a study by Taguchi et al. [16], it was reported that 18.2% and 37.8% MHCI were found in TP and SC muscles, respectively, by myosin-ATPase staining, whereas another report showed no detection of MHCI in any pharyngeal muscles [17]. These discrepancies are

Table III. Summary of SERCA1, SERCA2, and hybrid (S1+S2) expression values (mean ± SD) in humans and the results of unpaired t-tests (p-value).

	SERCA1	SERCA2	Hybrid (S1+S2)
Thyropharyngus (TP)			
Mean ± SD	74 ± 10.1	37.5 ± 11.8	9.5 ± 3.9
Cricopharyngus (CP)			
Mean ± SD	47.7 ± 14.5	63 ±13.1	16.2 ± 4.1
TP vs CP (P value)	< 0.05	< 0.05	< 0.05

P < 0.05 was considered as statistically significant; SD: standard deviation.

potentially derived from different techniques and a lack of consideration of the anatomical differences in rat hypopharyngeal muscles in which the SC muscle is likely present instead of CP muscle in upper species including humans (Fig. 1).

Our data of MHC and SERCA expression patterns demonstrated the presence of three different sub-classifications in pharyngeal muscle fibres: fast fibres represented by expression of SERCA1 and MHCII, which are associated with short bursts for rapid and powerful motion despite the lack of endurance; slow fibres represented by expression of SERCA2 and MHCI, which generate fatigue-resistant and sustainable motion with low peak power; hybrid intermediate fibres represented by MHCII, SERCA1, and SERCA2, which have a hybrid phenotype of fast and slow fibres i.e. fast and fatigue-resistant. In fact, muscle regions with a high proportion of fast fibres facilitate rapid, phasic and forceful movements as fast peristaltic contraction that propels the food bolus during swallow. Slow muscle fibres might play a role in maintenance of upper airway patency and prevent the pharyngeal tube from collapse with inspiratory negative pressure. Moreover, hybrid fibres may permit the muscle to recruit the desired form of contraction and relaxation, which allows highly specific fine-tuned movement. This complexity enables organisation of heterogeneous and simultaneous functions during sphincter tonic contraction and relaxation. In addition, the co-existence of both SERCA1 and 2 in either fibre type may be advantageous for more Ca^{2+}-handling versatility [18]. These observations support the hypothesis that the proportions and distribution patterns of MHC and SERCA in muscle fibres are closely related to unique and specific pharyngeal muscle functions.

In this study, we observed interesting histological features in the SC muscle in which fast and slow fibres appeared to divide the muscles into two indistinct layers: a fast-outer layer (FOL) containing MHCII/SERCA1 fibres and a slow inner layer (SIL) containing a higher percentage of SERCA2, MHCI, and hybrid fibres (Fig. 3). This property may allow them to function independently and supports the idea that both layers cooperatively generate a shearing effect in the UES. These results are compatible with previous reports indicating that the inner muscle layer with neonatal MHC myofibers is functionally analogous to the slow twitch myofiber layer in humans. Previous studies reported that the human CP, which resembles rat SC muscle, can also be divided into FOL and SIL fibres [19-21]. Mu et al. [21] suggested that the different distributions in SIL and FOL are reflective of their different physiological capabilities and independent functionalities. SIL is associated with fine movements and tonic activity, enabling sustained resting tone of the UES, which is required to prevent aerophagia and occasional forceful contraction during deglutition. FOL is associated with rapid and powerful contraction enabling prompt and phasic dynamics while swallowing. These findings suggest that both fibre

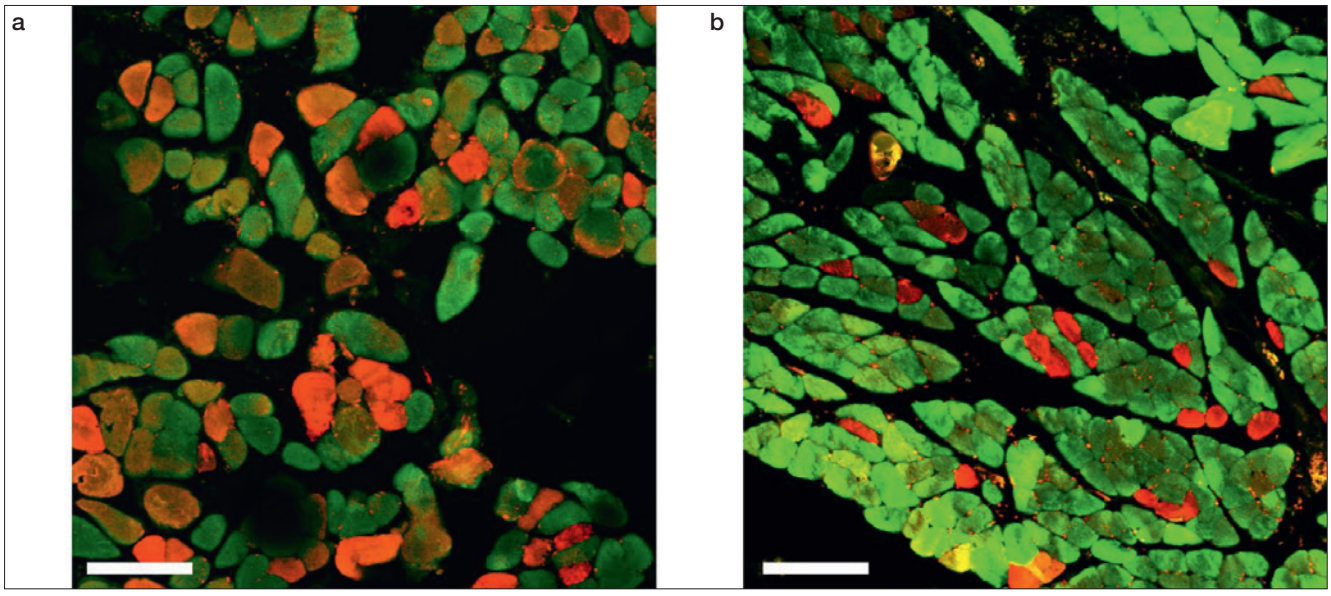

Fig. 5. SERCA isoform expression in human muscles: expression of both SERCA1 (green) and SERCA2 (red) were observed in the cricopharyngeus (CP) muscle (**a**) and thyropharyngeal (TP) muscle (**b**). SERCA1 was more predominant in the TP muscle, while SERCA2 was expressed more in the CP muscle. Scale bar = 50 μm.

layers have distinct contractile properties and play different roles.

Because muscles are generally susceptible to degeneration- and contraction-induced injuries associated with daily activities, which lead to muscle atrophy and decreased functionality with aging [22], we investigated the potential aging effects on expression of MHC and SERCA by comparative analysis between young and aged rats. The effect of aging was associated with maintenance of a higher proportion of fast MHCII/SERCA1 fibres. There were also significant increases in SERCA2 and hybrid fibres in all pharyngeal muscles. The age-associated shifts in MHC expression with decrease in MHCI support a muscle-specific change rather than follow a "fast to slow" rule. These changes may be associated with slowing of the contraction-relaxation cycle in pharyngeal muscles. Senescence is associated with loss of swallowing efficiency by slowing of the swallow sensitivity response in the pharynx, coordination of swallowing, impaired opening of the UES and loss of UES elasticity or compliance [23].

In humans, both SERCA1 and 2 were detected in TP and CP muscles. Notably, the frequency of slow muscle fibres (SERCA2) in CP muscle was significantly higher than that in TP muscle. This finding is supported by previous reports showing the predominance of MHCI slow twitch fibres in CP muscle (> 80%) [24 25]. Our study also showed that CP muscle had a significantly higher ratio of hybrid fibres than TP muscle. In a similar context, rat SC muscle, which is an analogue to CP muscle in humans, had the highest frequency of hybrid fibres (SERCA1 and SERCA2) among all examined muscles. These findings support the notion that CP muscles are relatively biased toward slow and hybrid fibres rather than fast fibres. In fact, TP muscle serves as an inferior pharyngeal constrictor and plays an important role in respiration and maintaining patency of the collapsible pharyngeal airway during swallowing, respiration and vocalisation, which can benefit from the predominance of fast fibres. In contrast, CP/SC muscles form a transition sphincter between the pharynx and oesophagus, whereby successive contractions of the pharyngeal constrictors during swallowing propel the bolus downward to the CP/SC muscles, followed by its relaxation to allow bolus passage into the oesophagus. Thus, MHCI and SERCA2 myofibers provide the tonic force to maintain closure of the UES, which benefits from the predominance of slow and hybrid fibres. In support of this speculation, slowing of peristalsis and decreased relaxation in the sphincter produce swallowing abnormalities, leading to retained food or saliva in the pharynx after swallowing [26]. Moreover, the variety of hybrid CP/SC muscle fibre types may explain its wide range of contraction characteristics. These observations support that differential compositions of fibre types are associated with differential motor functions of pharyngeal muscles, contributing to coordinated regulation for speech and swallowing movements.

Conclusions

In this study, we demonstrated different proportions and patterns of slow and fast muscle fibres represented by MHC and SERCA isoforms in pharyngeal muscles in humans and rat. Based on rigorous examination and comparative analyses of the obtained results, the following conclusions can be drawn:

- Pharyngeal muscles have different proportions and patterns of MHC and SERCA, which are potentially associated with coordination of pharyngeal muscle functions.
- Histological analysis of rat SC muscle revealed two distinct layers: SIL and FOL.
- Aging might be associated with an increase of SERCA2 and hybrid fibres in pharyngeal muscles.
- In humans, CP muscle has higher portions of slow muscle (SERCA2) and hybrid (SERCA1 and 2) fibres in comparison with TP muscles.

These observations may provide a new insight into further physiology for regulation of pharyngeal movement and function.

Acknowledgements

This work was supported by a Grant-in-Aid for Scientific Research (15K1082, 25293350, and 17H07016) from the Japan Society for the Promotion of Science.

Conflict of interest statement

None declared.

References

1 Mu L, Sanders I. *Neuromuscular compartments and fiber-type regionalization in the human inferior pharyngeal constrictor muscle.* Anat Rec 2001;264:367-77. https://doi.org/10.1002/ar.10020.

2 Cunningham ET, Jones B. *Anatomical and physiological overview.* In: Jones B, editor. *Normal and abnormal swallowing.* New York, NY: Springer; 2003. pp. 11-34.

3 Mittal RK. *Motor function of the pharynx, the esophagus, and its sphincters.* In: *Physiology of the gastrointestinal tract*. Fifth edition. San Rafael (CA): Morgan & Claypool Life Sciences; 2011. pp. 919-50.

4 Khan A, Carmona R, Traube M. *Dysphagia in the elderly*. Clin Geriatr Med 2014;30:43-53. https://doi.org/10.1016/j.cger.2013.10.009.

5 Molfenter SM, Steele CM. *The relationship between residue and aspiration on the subsequent swallow: an application of the normal-*

ized residue ratio scale. Dysphagia 2013;28:494-500. https://doi.org/10.1007/s00455-013-9459-8.

6 Thomas LB. *Effect of dystrophin deficiency on selected intrinsic laryngeal muscles of the mdx mouse.* University of Kentucky Doctoral Dissertations 2008;591.

7 Vinney LA, Connor NP. *Structure and function of the laryngeal and pharyngeal muscles.* In: McLoon LK, Andrade F, editors. *Craniofacial muscles.* New York, NY: Springer; 2012. pp. 141-66.

8 Kjellgren D, Ryan M, Ohlendieck K, et al. *Sarco(endo) plasmic reticulum Ca2+ ATPases (SERCA1 and-2) in human extraocular muscles.* Invest Ophthalmol Vis Sci 2003;44:5057-62. https://doi.org/10.1167/iovs.03-0218.

9 Lipskaia L, Chemaly ER, Hadri L, et al. *Sarcoplasmic reticulum Ca2+ ATPase as a therapeutic target for heart failure.* Expert Opin Biol Ther 2010;10:29-41. https://doi.org/10.1517/14712590903321462.

10 Periasamy M, Kalyanasundaram A. *SERCA pump isoforms: their role in calcium transport and disease.* Muscle Nerve 2007;35:430-42. https://doi.org/10.1002/mus.20745.

11 Hoh JFY. *Laryngeal muscle fibre types.* Acta Physiol 2005;183:133-49. https://doi.org/10.1111/j.1365-201X.2004.01402.x.

12 Mu L, Sanders I. *Newly revealed cricothyropharyngeus muscle in the human laryngopharynx.* Anat Rec 2008;291:927-38. https://doi.org/10.1002/ar.20727.

13 Kobler JB, Datta S, Goyal RK, et al. *Innervation of the larynx, pharynx, and upper esophageal sphincter of the rat.* J Comp Neurol 1994;349:129-47. https://doi.org/10.1002/cne.903490109.

14 Whitmore I. *Oesophageal striated muscle arrangement and histochemical fibre types in guinea-pig, marmoset, macaque and man.* J Anat 1982;134:685-95.

15 Bonington A, Whitmore I, Mahon M. *A histological and histochemical study of the cricopharyngeus muscle in the guinea-pig.* J Anat 1987;153:151-61.

16 Taguchi A, Hyodo M, Yamagata T, et al. *Age-related remodeling of the hypopharyngeal constrictor muscle and its subneural apparatuses: a scanning electron microscopical study in rats.* Dysphagia 2004;19:241-7.

17 Randolph ME, Luo Q, Ho J, et al. *Ageing and muscular dystrophy differentially affect murine pharyngeal muscles in a region-dependent manner.* J Physiol 2014;592:5301-15. https://doi.org/10.1113/jphysiol.2014.280420.

18 Fajardo VA, Bombardier E, Vigna C, et al. *Co-expression of SERCA isoforms, phospholamban and sarcolipin in human skeletal muscle fibers.* PLoS One 2013;12:e84304. https://doi.org/10.1371/journal.pone.0084304.

19 Mu L, Sanders I. *Muscle fiber-type distribution pattern in the human cricopharyngeus muscle.* Dysphagia 2002;17:87-96. https://doi.org/10.1007/s00455-001-0108-2.

20 Mu L, Sanders I. *Neuromuscular specializations within human pharyngeal constrictor muscles.* Ann Otol Rhinol Laryngol 2007;116:604-17. https://doi.org/10.1177/000348940711600809.

21 Mu L, Su H, Wang J, et al. *Myosin heavy chain-based fiber types in the adult human cricopharyngeus muscle.* Muscle Nerve 2007;35:637-48. https://doi.org/10.1002/mus.20741.

22 Faulkner JA, Brooks SV, Zerba E, et al. *Muscle atrophy and weakness with aging: contraction-induced injury as an underlying mechanism.* J Gerontol A Biol Sci Med Sci 1995;50 Spec No:124-9. https://doi.org/10.1093/gerona/50a.special_issue.124.

23 McKee GJ, Johnston BT, McBride GB, et al. *Does age or sex affect pharyngeal swallowing?* Clin Otolaryngol Allied Sci 1998;23:100-6.

24 Bonington A, Mahon M, Whitmore I. *A histological and histochemical study of the cricopharyngeus muscle in man.* J Anat 1988;156:27-37.

25 Kristmundsdottir F, Mahon M, Froes MMQ, et al. *Histomorphometric and histopathological study of the human cricopharyngeus muscle.* Neuropathol Appl Neurobiol 1990;16:461-75.

26 Mu L, Sobotka S, Chen J, et al. *Beach TG Arizona Parkinson's Disease Consortium. Altered pharyngeal muscles in Parkinson disease.* J Neuropathol Exp Neurol 2012;71:520-30. https://doi.org/10.1097/NEN.0b013e318258381b.

Received: May 1, 2018 - Accepted: June 12, 2018

How to cite this article: Elrabie Ahmed M, Bando H, Fuse S, et al. *Differential isoform expression of SERCA and myosin heavy chain in hypopharyngeal muscles.* Acta Otorhinolaryngol Ital 2019;39:220-229. https://doi.org/10.14639/0392-100X-2185

Address for correspondence: Takahiro Tsujikawa, Department of Otolaryngology-Head and Neck Surgery, Kyoto Prefectural University of Medicine, 465 Kajiicho, Kamigyoku, Kyoto City, 602-0841 Kyoto, Japan. Tel. +81-75-251-5603. Fax +81-75-251-5604. E-mail: tu-ji@koto.kpu-m.ac.jp

ACTA OTORHINOLARYNGOLOGICA ITALICA 2019;39:230-234; doi: 10.14639/0392-100X-1576

HEAD AND NECK

Systemic inflammatory response after endoscopic surgery of Zenker's diverticulum

Risposta infiammatoria sistemica dopo trattamento chirurgico endoscopico del diverticolo di Zenker

S. HERING[1], S. WIEGAND[2]

[1] Department of Otorhinolaryngology, Head and Neck Surgery, University Hospital Giessen and Marburg GmbH, Marburg, Germany; [2] Department of Otorhinolaryngology, Head and Neck Surgery, University Hospital Leipzig, Leipzig, Germany

SUMMARY

Zenker's diverticulum can be treated with a carbon dioxide laser or linear stapling device. A retrospective study on patients undergoing elective surgery for Zenker`s diverticulum with carbon dioxide laser or stapler was performed to analyse possible differences in inflammatory responses during the postoperative period. Leucocyte counts and C-reactive protein levels in peripheral blood were measured before and on days 1, 2, 3 and 5 after the operation. Statistical analysis was performed using the Mann-Whitney U-test. Of 34 patients, 16 were treated by laser and 18 by stapler. Age, sex ratio and ASA grade did not differ between the groups. Postoperative leukocytosis was significantly milder in the stapler group compared with patients who were treated by carbon dioxide laser. The mean C-reactive protein (CRP) level on day 1, 2 and 3 after surgery was significantly higher in the CO_2 laser group than in the stapler group. Leukocyte counts recovered on day 3 after surgery in both groups while CRP levels did not decline to preoperative levels at day 5 after the operation in either group. No inflammatory complications such as mediastinitis or pneumonia occurred. In conclusion, the inflammatory response in the early period after carbon dioxide laser diverticulotomy of Zenker's diverticulum is higher than after stapler-assisted surgery of Zenker's diverticulum.

KEY WORDS: Zenker's diverticulum • C reactive protein • Leukocytosis • Stapler • CO2 laser

RIASSUNTO

Il diverticolo di Zenker può essere trattato con il laser CO2 o con l'utilizzo di una suturatrice lineare. È stato quindi realizzato questo studio retrospettivo condotto su pazienti affetti da diverticolo di Zenker sottoposti, in elezione, a trattamento chirurgico o con laser CO2 o con suturatrice al fine di analizzare eventuali differenze nella risposta infiammatoria durante il periodo post-operatorio. Sono state misurate la conta dei leucociti e la proteina C-reattiva nel sangue periferico sia il giorno prima dell'operazione, sia in prima, seconda, terza e quinta giornata post-operatoria. È stata quindi eseguita l'analisi statistica utilizzando il Test U di Mann-Whitney. Dei 34 pazienti, 16 sono stati trattati con laser e 18 con suturatrice. Non c'erano differenze di età, sesso e grado ASA tra i due gruppi. La leucocitosi postoperatoria è stata significativamente più modesta nei pazienti trattati con suturatrice rispetto a quelli trattati con laser, e i livelli di proteina C-reattiva in prima, seconda e terza giornata post-operatoria si sono rivelati significativamente più alti nei pazienti trattati con laser. La conta dei leucociti si è normalizzata in terza giornata postoperatoria, invece i livelli di proteina C-reattiva, in entrambi i gruppi, non sono diminuiti, neppure in quinta giornata postoperatoria. Non si sono verificate complicanze infiammatorie, quali polmoniti o mediastiniti. In conclusione, la risposta infiammatoria precoce dopo diverticolomia con laser CO_2 di un diverticolo di Zenker è maggiore rispetto a quella dopo diverticolomia tramite suturatrice.

PAROLE CHIAVE: *Diverticolo di Zenker • Proteina C reattiva • Leucocitosi • Suturatrice • Laser CO2*

Introduction

Zenker's diverticulum is an acquired pulsion diverticulum of the mucosa which develops in the so-called Killian triangle, a triangular shaped area between the oblique muscle fibres of the inferior pharyngeal constrictor muscle and the horizontal muscle fibres of the cricopharyngeus muscle. Zenker's diverticula typically present in middle-aged and elderly individuals, especially during the seventh and eighth decades of life. Dysphagia is the main presenting symptom; other symptoms are regurgitation of undigested food, choking, cough, halitosis, globus, aspiration, pneumonia and oesophageal obstruction.

Zenker's diverticulum was first reported in 1767 by Ludlow [1], but named after the German pathologist Friedrich Albert von Zenker who published a case series in 1877 [2]. Since the first report of successful open surgical treatment

by Wheeler in 1886 [3], many options have been described for the treatment of Zenker's diverticulum. The external approach was the treatment of choice for many years. Mosher in 1917 introduced the treatment by rigid endoscopy, sectioning the septum of the diverticulum [4]. In the last decades, endoscopic treatment has gained more popularity also due to the use of carbon dioxide laser, which was introduced by van Overbeek et al. [5]. In 1993, Collard at al. introduced diverticulotomy by using the endostapler [6].

Since the invention of the carbon dioxide laser and the linear stapling device, these methods have become widely used once they were demonstrated to be secure and precise [7-9]. Compared with classic surgery the main advantages of the endoscopic approach are shorter operative time, less surgical trauma and lower morbidity rate [10].

Until now, it is unclear which technique is superior in terms of outcomes. It is controversial if the open wound after laser surgery leads to bacterial penetration into the mediastinum and if closure of the wound by stapler prevents a mediastinal bacterial invasion, and therefore minimises complication rates like mediastinitis. Therefore, the issue of early diagnosis of inflammatory changes in the post-operative period is important. Post-operative serial screening of white blood cell count (WBC) and C-reactive protein (CRP) is useful as an indicator of infectious complications. CRP is an acute-phase protein synthesised by hepatocytes, largely in response to pro-inflammatory cytokines [11]. CRP is not specific for a particular disease, but can arise due to inflammation, trauma, malignancy and tissue infarction. In bacterial infection, CRP levels increase within 6 hours [12]. CRP is considered to be useful for detection of an inflammatory response early in its course, and also for monitoring disease activity [11]. Leukocytosis also commonly accompanies infection and may serve as an early marker for a developing infection. Normal WBC and CRP responses can rule out almost all early infectious complications.

There is no study that has investigated inflammatory changes after rigid endoscopic surgery of Zenker's diverticulum by laser or stapler. The objective of the present study was to investigate whether endoscopic stapler surgery causes a less pronounced release of inflammatory cytokines compared to endoscopic laser surgery.

Materials and methods

This retrospective study included patients having elective endoscopic surgery for Zenker's diverticulum by carbon dioxide laser or stapler from 2001 to 2011 and fulfilled the following inclusion criteria:

- The diagnosis of Zenker's diverticulum was made through clinical and radiologic examination.
- Patients had a baseline examination 1 or 2 days before the procedure. Blood samples for biochemistry were obtained as part of the preoperative examination (day 0) and subsequently at day 1, 2, 3 and 5 upon completion of the procedure.
- C-reactive protein (CRP) and white blood cell count (WBC) at baseline were in the normal range (CRP < 5 mg/l, WBC 4000-10,000/µl). Therefore, any inflammatory situations due to comorbidity could be excluded.

The procedures were performed under general anaesthesia. Patients were placed supine with their neck overextended. The diverticular sac was exposed with a Weerda distending operation laryngoscope. After transoral positioning of the laryngoscope in the hypopharynx, the wall containing cricopharyngeus muscle fibers between the diverticular sac and hypopharynx lumen was visualised. Then, division of the septum was performed by CO_2 laser or stapler. By endoscopically dividing the cricopharyngeal muscle the diverticula were marsupialised to become one cavity with the hypopharynx. Post-operatively, patients were kept on total parenteral nutrition for a period of at least 48 hours. A liquid oral diet was introduced at first, followed by solids according to the patients' acceptance. No post-operative routine radiological swallow examination was performed. All patients received intravenous fluid infusion, peri- and post-operative intravenous antibiotics [cefuroxime (1.5 g per 8 h) and metronidazole (500 mg per 12 h)], a proton pump inhibitor and analgesics.

The ASA score is a subjective assessment of a patient's overall health that is based on six classes (I to VI). I: A normal healthy patient; II: A patient with mild systemic disease; III: A patient with severe systemic disease; IV: A patient with severe systemic disease that is a constant threat to life; V: A moribund patient who is not expected to survive without the operation; VI: A declared brain-dead patient whose organs are being removed for donor purposes [13].

Statistical analysis

The Mann-Whitney-U-test was used to compare sex, age, ASA grade, and WBC and CRP levels of both patient groups. Results were considered statistically significant if $p \leq 0.05$.

Results

In all, 34 patients fulfilled the inclusion criteria and were investigated. Of these, 18 patients underwent stapler-assisted diverticulotomy, and the remaining 16 patients were

subjected to diverticulotomy by carbon dioxide laser. All procedures were performed by an experienced surgeon, who was defined as one having more than 5 years experience as an attending surgeon. In each group, 3 patients were endoscopically treated for Zenker's diverticulum in other hospitals before. Basic parameters of both patient groups prior to Zenker's diverticulum surgery are shown in Table I. There was no significant difference between the two groups of patients in terms of age, sex ratio and ASA grade.

The mean operative time was 15 minutes shorter in the stapler group (53 vs 68 minutes); however, this difference was not statistically significant. No serious post-operative complications such as mediastinitis or pneumonia occurred in either group.

Before the operation, the levels of neither CRP nor WBC were significantly different between two groups. In both groups the WBC peaked as early as one day post-surgery, returning to normal range at the third day after surgery in all patients of both groups. However, there was a significant difference on the first day after surgery between the groups regarding the mean WBC. The patient group who was subjected to carbon dioxide laser diverticulotomy had a significant higher WBC than patients after stapler-assisted surgery ($p < 0.05$) (Fig. 1).

Mean CRP levels reached their maximum 2 days after surgery in both groups and did not decline to preoperative levels at day 5 after the operation. The increase in the carbon dioxide laser group was significantly higher than in the stapler group. Significant differences between both groups were seen on day 1, 2 and 3 after surgery ($p < 0.05$). Figure 2 shows the chronological change in the mean CRP level after surgery.

In one patient of the stapler group revision surgery was performed 6 months later due to recurrence of the diverticulum. In the other patients, no recurrence was observed.

Table I. Demographic details of patients.

	Stapler	Laser
Patients	18	16
Gender		
Male	11	9
Female	7	7
Age mean (range)	68.4 (46-86)	70.9 (56-85)
ASA grade (mean)	2.4	2.5
Primary surgery	15	13
Revision surgery	3	3

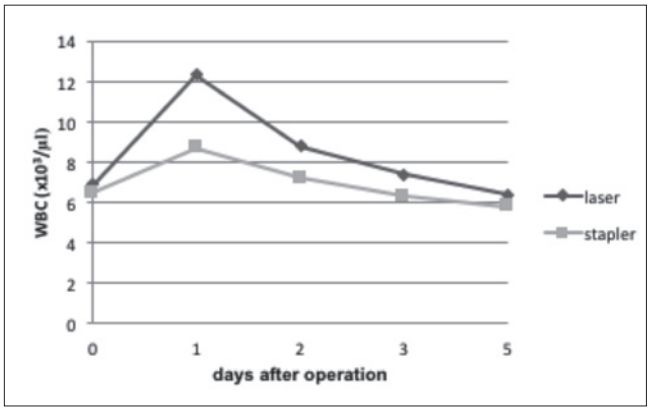

Fig. 1. WBC at baseline (0) and days 1, 2, 3 and 5 after surgery (mean values) in the carbon dioxide laser and stapler groups.

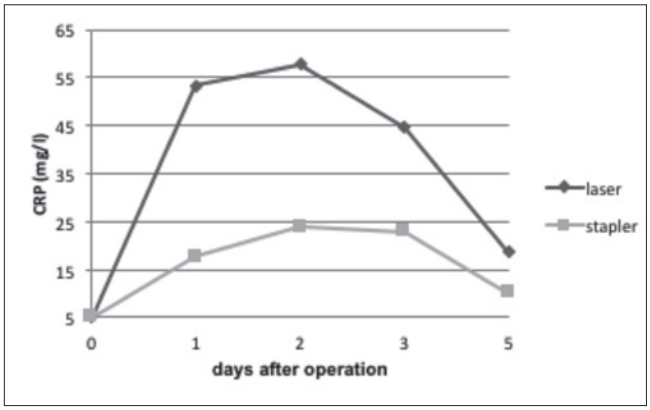

Fig. 2. CRP at baseline (0) and days 1, 2, 3 and 5 after surgery (mean values) in the carbon dioxide laser and stapler groups.

Discussion

Due to its good safety and long-term efficacy, rigid endoscopic surgery of Zenker's diverticulum has become increasingly popular, although there is some controversy about the optimal surgical method [14-16]. A recent meta-analysis demonstrated increased non-dental complications in patients treated by CO_2 laser compared to patients treated by stapler, but no difference in overall complications or revision surgery [17]. Endoscopic surgery for Zenker's diverticulum can be complicated by inflammatory processes and mediastinitis [18]. An elevated WBC and elevated CRP have been associated with an increased risk of inflammatory events. Until now, there are no studies on the inflammatory response after laser surgery or stapler surgery for Zenker's diverticulum. Therefore, the influence of carbon dioxide laser surgery and stapler surgery of Zenker's diverticulum on WBC and CRP were retrospectively investigated.

The present study shows no differences in pre-operative clinical parameters between the two groups. The results showed that endoscopic surgery for Zenker's diverticulum whether performed by carbon dioxide laser or stapler is associated with an inflammatory response. There were no differences between CRP and WBC count at baseline between both groups since values within the normal range at baseline were defined as inclusion criteria. The stapler group was superior to the carbon dioxide laser group in terms of post-operative systemic inflammation. There was a significant lower mean WBC on the first post-operative day and significant lower CRP levels on days 1, 2 and 3 after surgery in patients treated by stapler.

The reason for the lower inflammatory response in the stapler group may is that carbon dioxide laser diverticulotomy causes greater tissue trauma since the stapler has the advantage of simultaneously cutting and sealing the wound edge. It is possible that the inflammatory reactions were the result of possible unnoticed leakages. In order for a leak to occur, the peri-oesophageal fascia must be violated. Because the fascial planes in the neck are continuous with the mediastinum, leakage of oesophageal contents into the neck can cause mediastinitis. The greater physical trauma due to laser surgery may lead to an increased liberation of inflammatory cytokines and enhancing post-operative systemic inflammation. Another cause for the higher inflammatory response in the CO_2 laser group could be the longer mean operative time which was 15 minutes longer in the CO_2 laser group. This difference was not statistically significant although the longer duration of tissue injury can lead to a greater amount of released mediators resulting in a higher inflammatory response. However, in our patients no inflammatory complications occurred and there were no significant differences in outcomes. In one patient in the stapler group a recurrence of the diverticulum occurred, while in the laser group there were no recurrences.

There are some limitations to our study. The first and most obvious is that it consists of a relatively small number of patients who were studied retrospectively. We know that laser surgery is possible when stapler surgery is not, for example in cases with cricopharyngeal dysfunction or small diverticulum. Due to the retrospective nature of the study, we do not know what influences the decision for stapler or laser surgery. Additionally, all patients were treated with intravenous antibiotics in the post-operative period since these antibiotics were part of our treatment scheme. This affected post-operative inflammatory reaction, although antibiotic treatment was the same in both groups.

Conclusions

The inflammatory response in the early period after carbon dioxide laser diverticulotomy of Zenker's diverticulum is higher than after stapler-assisted surgery of Zenker's diverticulum. Prospective studies with larger numbers of patients are needed to further address this issue.

Conflict of interest statement

None declared.

References

1 Ludlow A. *A case of obstructed deglutition from a preternatural dilatation of and bag formed in the pharynx*. Med Obs Inq 1767;3:85.

2 Zenker FA, von Ziemssen H. *Krankheiten des Oesophagus.* In: Von Ziemssen H, ed. *Handbuch der speziellen Pathologie und Therapie*. Leipzig, Germany: FC Vogel; 1877. pp. 1-87.

3 Wheeler WI. *Pharyngocele and dilatation of the pharynx, with existing diverticulum at lower portion of pharynx lying posterior to the esophagus, cured by pharyngotomy, being the first of the time recorded*. Dublin J Med Science 1886;82:349-56.

4 Mosher HP. *Webs and pouches of the oesophagus, their diagnosis and treatment.* Surg Gynecol Obstet 1917;25:175-87.

5 von Overbeek JJM, Hoeksma P, Edens ETH. *Microendoscopic surgery of the hypopharyngeal diverticulum using electrocoagulation or carbon dioxide laser.* Ann Otol Rhinol Laryngol 1984;93:34-6. https://doi.org/10.1177/000348948409300108.

6 Collard JM, Otte JB, Kenstens PJ. *Endoscopic stapling technique of esophago-diverticulostomy for Zenker's diverticulum.* Ann Thorac Surg 1993;56:573-6. https://doi.org/10.1016/0003-4975(93)90906-x.

7 Wilken R, Whited C, Scher RL. *Endoscopic staple diverticulostomy for Zenker's diverticulum: review of experience in 337 cases*. Ann Otol Rhinol Laryngol 2015;124:21-9. https://doi.org/10.1177/0003489414542421.

8 Anagiotos A, Feyka M, Eslick GD, et al. *Long-term symptom control after endoscopic laser-assisted diverticulotomy of Zenker's diverticulum*. Auris Nasus Larynx 2014;41:568-71. https://doi.org/10.1016/j.anl.2014.06.002.

9 Pollei TR, Hinni ML, Hayden RE, et al. *Comparison of carbon dioxide laser-assisted versus stapler-assisted endoscopic cricopharyngeal myotomy*. Ann Otol Rhinol Laryngol 2013;122:568-74. https://doi.org/10.1177/000348941312200906.

10 Koch M, Mantsopoulos K, Velegrakis S, et al. *Endoscopic laser-assisted diverticulotomy versus open surgical approach in the treatment of Zenker's diverticulum*. Laryngoscope 2011;121:2090-4. https://doi.org/10.1002/lary.22152.

11 Cole DS, Watts A, Scott-Coombes D, et al. *Clinical utility of peri-operative C-reactive protein testing in general surgery.* Ann R Coll Surg Engl 2008;90:317-21. https://doi.org/10.1308/003588408X285865.

12 Pepys MB. *C-reactive protein fifty years on.* Lancet 1981;1:653-7. https://doi.org/10.1016/s0140-6736(81)91565-8.

13 https://www.*asahq*.org/resources/clinical-information/asa-physical-status-classification-system

14 Verhaegen VJ, Feuth T, van den Hoogen FJ, et al. *Endoscopic carbon dioxide laser diverticulostomy versus endoscopic staple-assisted diverticulostomy to treat Zenker's diverticulum*. Head Neck 2011;33:154-9. https://doi.org/10.1002/hed.21413.

[15] Adam SI, Paskhover B, Sasaki CT. *Laser versus stapler: outcomes in endoscopic repair of Zenker diverticulum*. Laryngoscope 2012;122:1961-6. https://doi.org/10.1002/lary.23398.

[16] Seth R, Rajasekaran K, Lee WT, et al. *Patient reported outcomes in endoscopic and open transcervical treatment for Zenker's diverticulum*. Laryngoscope 2014;124:119-25. https://doi.org/10.1002/lary.24152.

[17] Parker NP, Mison S. *Carbon dioxide laser versus stapler-assisted endoscopic Zenker's diverticulotomy: a systematic review and meta-analysis*. Otolaryngol Head Neck Surg 2014;150:750-3. https://doi.org/10.1177/0194599814521554.

[18] Helmstaedter V, Engel A, Hüttenbrink KB, et al. *Carbon dioxide laser endoscopic diverticulotomy for Zenker's diverticulum: results and complications in a consecutive series of 40 patients.* ORL J Otorhinolaryngol Relat Spec 2009;71:40-4. https://doi.org/10.1159/000170379.

Received: January 16, 2017 - Accepted: December 1, 2017

How to cite this article: Hering S, Wiegand S. *Systemic inflammatory response after endoscopic surgery of Zenker's diverticulum.* Acta Otorhinolaryngol Ital 2019;39:230-234. https://doi.org/10.14639/0392-100X-1576

Address for correspondence: Susanne Wiegand, Department of Otolaryngology, Head and Neck Surgery, University Hospital Leipzig, Liebigstrasse 12, 04103 Leipzig, Germany. Tel. +49 0341 9721945. E-mail: susanne.wiegand@medizin.uni-leipzig.de

HEAD AND NECK

Patterns of recurrence after open partial horizontal laryngectomy types II and III: univariate and logistic regression analysis of risk factors

Patterns di recidiva dopo intervento di laringectomia parziale orizzontale tipo II e III: analisi univariata e regressione logistica dei fattori di rischio

E. CROSETTI[1], A. BERTOLIN[2], G. MOLTENI[3], I. BERTOTTO[4], D. BALMATIVOLA[5], M. CARRARO[1], A.E. SPRIO[6], G.N. BERTA[6], L. PRESUTTI[7], G. RIZZOTTO[2], G. SUCCO[1, 8]

[1] Head and Neck Oncology Service, Candiolo Cancer Institute - FPO IRCCS, Candiolo (TO), Italy; [2] Otolaryngology Service, Vittorio Veneto Hospital, Vittorio Veneto (TV) Italy; [3] Department of Otolaryngology-Head and Neck Surgery, University Hospital of Verona, Italy; [4] Radiology Service, Candiolo Cancer Institute - FPO IRCCS, Candiolo (TO), Italy; [5] Pathology Service, Candiolo Cancer Institute - FPO IRCCS, Candiolo (TO), Italy; [6] Department of Clinical and Biological Sciences, University of Turin, Italy; [7] Otolaryngology Service, Head and Neck Dept., Policlinico Hospital, University of Modena, Italy; [8] Oncology Dept. University of Turin, Italy

SUMMARY

In choosing the best surgical treatment (total or partial laryngectomy) for patients affected by laryngeal squamous cell carcinoma (SCC), it is still necessary to identify a link between prognostic factors and oncological outcomes. A retrospective analysis of clinical outcomes of 819 patients affected by laryngeal cancer who underwent OPHL type II and III between 1995 to 2014 was carried out. Focusing on recurrence and its site (local, regional or distant), our cohort has been divided in two groups: patients showing recurrence (n = 108) vs those without recurrence (n = 711). Thirteen clinical-pathological parameters have been studied by univariate and multivariate analysis to identify possible correlations between recurrence and oncological outcomes (overall survival (OS), disease free survival (DFS), disease specific survival (DSS), laryngectomy free survival (LSF), laryngectomy free freedom (FFL). In multivariate analysis, we found 4 negative prognostic factors for recurrence: site of tumour (> supraglottic), cartilage invasion (> if present), perineural invasion (> if present) and type of OPHL (> in OPHL type III). The knowledge and detection of negative prognostic factors for the risk of recurrence (pN classification, cartilage involvement, perineural invasion, and thus the type of surgical treatment adopted) could increase the already well-established potentiality of OPHLs in treating cases with a safe indication after careful discussion in the tumour board.

KEY WORDS: Open partial horizontal laryngectomy • Supracricoid partial laryngectomy • Supratracheal partial laryngectomy • Recurrence • Laryngeal cancer • Multivariate analysis

RIASSUNTO

Nella scelta del trattamento chirurgico (laringectomia totale o parziale) in pazienti affetti da carcinoma laringeo, è ancora necessaria l'identificazione di una correlazione tra fattori prognostici e risultati oncologici. Una coorte multi-istituzionale di 819 pazienti affetti da carcinoma laringeo e sottoposti a laringectomia parziale orizzontale (OPHL) tipo II e tipo III dal 1995 al 2014 è stata suddivisa in base alla comparsa o meno di recidiva (108 vs 711) ed alla sede di ricomparsa di malattia (loco-regionale, locale, a distanza). Sono state analizzate, mediante analisi uni e multivariata, tredici variabili cliniche e istologiche nei due gruppi, in relazione agli esiti oncologici (OS, DFS, DSS, LSF, FFL). All'analisi multivariata quattro sono risultati i fattori determinanti rischio di recidiva: sede del tumore (maggiormente nei tumori sopraglottici), invasione cartilaginea (maggiormente se presente), invasione perineurale (maggiormente se presente) ed il tipo di OPHL (maggiormente in caso di OPHL tipo III). La conoscenza e la detezione dei fattori prognostici negativi per il rischio di recidiva (classificazione pN, interessamento cartilagineo, invasione perineurale e quindi trattamento chirurgico adottato) potrebbero aumentare le già note potenzialità delle OPHL nel trattare casi con indicazione certa dopo attenta discussione collegiale.

PAROLE CHIAVE: *Laringectomia parziale orizzontale • Laringectomia parziale sopracricoidea • Laringectomia parziale sopratracheale • Recidiva • Carcinoma laringeo • Analisi multivariata*

Introduction

Nowadays, surgery can be offered to patients affected by laryngeal cancer as a valuable method to preserve part of the larynx and its functions, avoiding the negative physical and psychosocial impact of permanent tracheostomy [1 2]. In the last decade, organ-sparing techniques have been developed and extensively used, with excellent results, as upfront treatment in early [3 4] and in selected more advanced stages (cT3 and cT4) [5]. Different therapeutic options are now available for cT3 and cT4 larynx cancers [6 7] but a dichotomy exists considering locally advanced tumours: is the lesion amenable to partial laryngectomy or to total laryngectomy? During multidisciplinary discussions about larynx cancer treatment, this is not a simple question.

Each time, the surgeon faces the decision to attempt conservative treatment or to address the patient to a safer up-front total laryngectomy. The choice of treatment to recommend can be difficult to understand, especially for young or less experienced surgeons. In fact, if two lesions are at the same stage and patients are free from contraindications related to general conditions, why are they selected for different treatments?

This is because a clear consensus is still lacking for what concerns simple and objective parameters that should be used in the selection of patients amenable to one or the other technique. Although staging of laryngeal cancer has been considered for a long period the criterion that better correlates with loco-regional control and patient survival, prognosis is a complex phenomenon arising from tumour and patient characteristics, molecular biology, genetics and environment [8 9]. However, the importance of each has not been unequivocally demonstrated. To complicate matters further, negative prognostic factors involved in recurrence onset are not established universally, but are important to discriminate the most suitable treatment for the patient.

The application of logistic regression models to retrospective studies allows detection and/or analysis of prognostic factors that are potentially useful to predict the main oncologic and functional outcomes. Furthermore, data emerging from large multi-institutional series of cases who were treated with the same protocols provide insights about the most controversial aspects in the treatment indications.

In this multi-institutional retrospective study, we analysed a large series of locally advanced laryngeal cancer patients treated by open partial horizontal laryngectomies (OPHL) spurred by a dual aim: a) the identification of novel prognostic factors correlating with disease recurrence; b) improvements for more correct use of OPHL as a single and up-front laryngeal cancer treatment, based on results of the previous point.

Patients and methods

Patients

The study was conducted retrospectively analysing medical records from 819 patients affected by laryngeal cancer who underwent OPHL between January 1995 and December 2014 in tertiary reference Italian hospitals: Hospital of Vittorio Veneto (Treviso), Martini Hospital (Turin), Candiolo Cancer Institute (Turin) and Policlinico Hospital (Modena). Patient characteristics, distribution according to the involved laryngeal sites, as well as their pT and pN categories (2009 TNM classification system) [10], are reported in Table I.

Inclusion criteria were histological diagnosis of glottic or supraglottic laryngeal squamous cell carcinoma (LSCC), Karnofsky index [11] higher than 80 and amenability to OPHL type II for advanced laryngeal cancers maintaining laryngeal functions [12] and type III surgeries for glottic/transglottic cancers extending to the cricoid or to extralaryngeal space [1 13 14].

Exclusion criteria were: a) purely supraglottic T3 tumour with limited extension to the pre-epiglottic space (and therefore amenable to OPHL type I or transoral laser microsurgery); b) lesions extended to base of tongue or pyriform sinus; c) lesions with major invasion of pre-epiglottic space involving the hyoid bone, involving the inter-arytenoid space, the posterior commissure and both arytenoid cartilages; d) large extralaryngeal spread of cancer involving thyroid gland, strap muscles, cervical skin, internal jugular vein or common carotid artery; e) lesions reaching the first tracheal ring; f) severe diabetes mellitus; g) severe chronic obstructive pulmonary disease; h) neurological problems impairing the ability to expectorate and/or swallow; and i) severe cardiac disease. The presence of clinically positive nodes > cN1 was not considered as an absolute contraindication. However, it should not represent a good indication for OPHL due to the probable need for post-operative radiotherapy (RT) [15].

One hundred and forty patients (17.1%) had already been treated previously for laryngeal carcinoma by CO_2 transoral laser surgery (61/140; 43.6%), radiation therapy (39/140; 27.9%), laser surgery and radiation therapy (11/140; 7.8%), cordectomy (21/140; 15.0%), open partial laryngectomy (7/140; 5.0%), or chemotherapy (1/140; 0.7%).

Thirteen clinical-pathological parameters are studied as possible negative prognostic factors: previous treatments,

Table I. Epidemiologic and clinical characteristics of patients treated by OPHL in the present series (N = 819).

		No of patients (%)	
Age	Mean ± standard deviation	60.2 ± 9.4	
	Range	16-87	
Gender	Male	755 (92.2%)	
	Female	64 (7.8%)	
Arytenoid mobility	Normal	581 (70.9%)	
	Impaired/fixed	238 (29.1%)	
pTN (VII ed.)		**Glottic**	**Supraglottic**
pT2	N0	201 (31.8%)	22 (11.8%)
	N1	2 (0.3%)	4 (2.2%)
	N2	3 (0.5%)	4 (2.2%)
pT3	N0	291 (46.0%)	86 (46.2%)
	N1	13 (2.1%)	11 (6.0%)
	N2	13 (2.1%)	23 (12.4%)
pT4	N0	94 (14.7%)	27 (14.4%)
	N1	10 (1.6%)	4 (2.2%)
	N2	6 (0.9%)	5 (2.6%)
Total		633 (77.3%)	186 (22.7%)

type of surgery, tumour site, pT subcategory, pN classification, grading, cartilage involvement, vascular invasion, perineural invasion, Delphian lymph node pN+, extranodal extension (ENE), status of margins, adjuvant treatments (Table II). Age was not considered among these factors because elderly patients affected by laryngeal cancer can be treated as younger patients [16].

Surgical procedures

Total laryngectomy and OPHL Type II are the most established solutions for intermediate-advanced stage laryngeal tumours affecting the glottis. OPHL Type III, instead, is amenable to glottic/transglottic tumours with subglottic extension reaching the cricoid and glottic/transglottic T4a lesions with extralaryngeal progression through the caudal end of the thyroid cartilage and/or through the cricothyroid membrane [1].

According to this, after informed consent had been obtained, all patients underwent OPHL types II-III [17] with curative intent: type IIa = 159/819 (19.4%), type IIa+ARY = 354/819 (43.2%), type IIb = 46/819 (5.6%), type IIb+ARY = 138/819 (16.9%), type IIIa = 10 (1.2%), type IIIa+CAU = 99/819 (12.1%), type IIIb = 5/819 (0.6%), type IIIb+CAU = 8/819 (1.0%) (Table III).

Neck dissection (ND), graded according to the American Academy of Otolaryngology – Head and Neck Surgery Foundation classification [18], was performed in 704 patients (85.9%), and was unilateral in 606 (86.1%) and bilateral in 98 (13.9%) cases. ND was elective (ND levels II-IV) in 634 cN0 (90.1%) and curative in 70 cN > 0 (9.9%) patients. Level VI or unilateral paratracheal lymph node clearance was added in 449 (63.7%) patients. No ND was performed in 115 cN0 (14.1%) cases.

Adjuvant treatment

Based on pathological findings, 95 (11.6%) patients were subjected to adjuvant treatment. Ninety-four (99.0%) were subjected to radiotherapy. The indications were: pN>1 (60 patients - 63.8%) and/or gross extralaryngeal extension (34 patients - 36.2%) with or without positive margins. A large volume encompassing the primary site and all draining lymph nodes were irradiated with a dose of up to 54 Gy. Regions at higher risk for malignant dissemination received a 12 Gy boost (total, 66 Gy; range, 62–68 Gy). Because of a higher risk of local recurrence [Delphian nodes pN+, pN+ with extracapsular spread (ECS), more extended pT4a tumours showing positive/close margins toward pre-laryngeal tissues, perineural invasion, or cartilage invasion], 52/95 (55.3%) patients received cisplatin (100 mg/m^2 on days 1, 22, and 43) concomitant with radiotherapy.

The remaining patient (1.0%) was subjected to chemotherapy alone, due to presence of distant metastasis.

Statistical methods

OS, DSS, DFS, FFL, LFS and laryngo-oesophageal dysfunction-free (LEDFS) [19] survivals were assessed by Kaplan-Meier curves. Log-rank (LR) and Gehan-Bres-

Table II. Stratification of patients (N = 819) according with the clinic-pathological parameters evaluated as possible risk factors for development of recurrences.

Clinic-pathological parameters	Recurrences		Patients
	Negative	Positive	
Previous treatment			
Positive	119 (16.7%)	21 (19.4%)	140 (17.1%)
Negative	592 (83.3%)	87 (80.6%)	679 (82.9%)
Type of surgery			
OPHL Type II	620 (87.2%)	77 (71.3%)	697 (85.1%)
OPHL Type III	91 (12.8%)	31 (28.7%)	122 (14.9%)
Tumour site			
Supraglottic	149 (21.0%)	32 (29.6%)	181 (22.1%)
Glottic	556 (78.2%)	73 (67.6%)	629 (76.8%)
Other	6 (0.8%)	3 (2.8%)	9 (1.1%)
pT stage			
pT1	3 (0.4%)	2 (1.9%)	5 (0.6%)
pT2	217 (30.5%)	17 (15.7%)	234 (28.6%)
pT3	385 (54.1%)	51 (47.2%)	436 (53.2%)
pT4	106 (14.9%)	38 (35.2%)	144 (17.6%)
pN stage			
pN0	645 (90.7%)	76 (70.4%)	721 (88.0%)
pN1	30 (4.2%)	13 (12.0%)	43 (5.3%)
pN2	36 (5.1%)	19 (17.6%)	55 (6.7%)
Grading			
Basaloid	10 (1.4%)	3 (2.8%)	13 (1.6%)
G1	179 (25.2%)	11 (10.2%)	190 (23.2%)
G2	326 (45.9%)	45 (41.7%)	371 (45.3%)
G3	196 (27.6%)	49 (45.4%)	245 (29.9%)
Cartilage involvement			
Positive	116 (16.3%)	50 (46.3%)	166 (20.3%)
Negative	595 (83.7%)	58 (53.7%)	653 (79.7%)
Vascular invasion			
Positive	194 (27.3%)	43 (39.8%)	237 (28.9%)
Negative	517 (72.7%)	65 (60.2%)	582 (71.1%)
Perineural invasion			
Positive	119 (16.7%)	39 (36.1%)	158 (19.3%)
Negative	592 (83.3%)	69 (63.9%)	661 (80.7%)
Delphian lymph node pN+			
Positive	19 (2.7%)	10 (9.3%)	29 (3.5%)
Negative	692 (97.3%)	98 (90.7%)	790 (96.5%)
Extranodal extension			
Positive	18 (2.5%)	11 (10.2%)	29 (3.5%)
Negative	693 (97.5%)	97 (89.8%)	790 (96.5%)
Status of margins			
Positive	55 (7.7%)	13 (12.0%)	68 (8.3%)
Close	29 (4.1%)	14 (13.0%)	43 (5.3%)
Negative	627 (88.2%)	81 (75.0%)	708 (86.4%)
Adjuvant treatment			
Positive	67 (9.4%)	28 (25.9%)	95 (11.6%)
Negative	644 (90.6%)	80 (74.1%)	724 (88.4%)

Table III. Treatments performed in the 819 patients.

Type of treatment	N (%)
OPHL	
IIa	159 (19.4%)
IIa + ARY	354 (43.2%)
IIb	46 (5.6%)
IIb + ARY	138 (16.9%)
IIIa	10 (1.2%)
IIIa + CAU	99 (12.1%)
IIIb	5 (0.6%)
IIIb + CAU	8 (1.0%)

ARY: arytenoid; CAU: crico-arytenoid unit.

low-Wilcoxon (GBW, for early events) tests were used to compare Kaplan-Meier estimates among the different subcategories.

The endpoints considered were obtained as the length of time from the date of diagnosis to: date of death (OS), date of death from disease (DSS), date of first recurrence (DFS), date of salvage laryngectomy (FFL), date of salvage total laryngectomy or date of death (LFS), date of salvage total laryngectomy or date of tracheostomy and/or PEG for functional reasons or date of death (LEDFS).

Univariate regression with collinearity analysis was used to evaluate independent risk factors (previous treatment, type of surgery, tumour site, pT stage, pN stage, grading, cartilage involvement, vascular or perineural invasion, Delphian lymph node pN+, ENE, status of margins, and adjuvant treatment). Variables were than included in logistic regression model. Kaplan-Meier curves, LR and GBW tests were performed with GraphPad Prism version 7.0a (GraphPad Software, San Diego, CA, USA), whereas univariate and multivariate regression analyses were performed with IBM® SPSS® Statistics version 24 (IBM Corp., Armonk, NY, USA). The incidences of relevant prognostic factors were analysed by a chi-squared test (PRIMIT-statistics for biomedical learning version 3.03). For all analyses, $p < 0.05$ was considered as the threshold for statistical significance.

Results

Pathology

Pathology reports showed close margins (< 2 mm on the specimen side) in 43 cases of 819 (5.3%) and positive margins (negative at frozen sections on the mucosa taken from the remaining larynx, but positive at definitive histopathologic examination on specimen) in 68 cases (8.3%). Positive or closed margins were associated with more advanced pT classification in both previously treat-

ed ($p < 0.05$) and untreated ($p < 0.001$) patients, while it correlated with the employment of the more aggressive OPHL type III only in previously treated patients ($p < 0.01$).

We detected 166 patients with cartilage invasion among the 582 (28.5%) who are affected by T3-T4 tumours (categories that could have cartilage invasion as definition). In particular, we detected the following pattern of cartilage invasion: 86/166 (51.8%) inner cortex of thyroid cartilage, 59/166 (35.5%) full thickness thyroid cartilage, 13/166 (7.8%) cricoid cartilage and 8/166 (4.9%) epiglottis cartilage. Among patients who experienced a recurrence, 50 (46.3%) had pathologic involvement of cartilages, the majority (44, 88.0%) at level of the thyroid cartilage. Furthermore, the development of recurrences and crico-arytenoid fixation did not correlate with pathological grading of lesions.

Finally, 746/819 patients (91.1%) had been staged as cN0. In contrast, 44 (5.9%) became pN+ after ND. Overall, lymph node metastases were detected in 98 (11.9%) patients, of whom 55 (56.1%) had multiple metastases.

Patterns of failure

Globally, 108 (13.2%) patients developed recurrences within 5 years after surgery. Thirty-eight patients (35.2%) displayed local recurrence, 25 patients (23.1%) regional and 8 (7.4%) distant. Twenty (18.5%) had local and regional recurrences, 6 (5.6%) were regional and distant and 4 (3.7%) local and distant recurrences. 7 patients (6.5%) developed both loco-regional and distant recurrences.

Among the 69 patients affected by local recurrence, 55 were endolaryngeal (79.7%), 9 extralaryngeal (13.0%) and 5 indeterminate (7.3%). The regional recurrences were detected on level VI in 19/58 patients (32.8%), whereas it affected levels II-V in the remaining 39 cases (67.2%). Among the latter, 34 (87.2%) recurrences were detected homolaterally, 3 contralaterally (7.7%) and 2 bilaterally (5.1%).

Recurrences were detected equally in patients who received previous treatments (21/140,15.0%) or not (87/679, 12.8%, $p = 0.576$). Furthermore, local recurrences displayed similar patterns between the two groups ($p = 0.917$). Otherwise, although the difference was not statistically significant ($p = 0.065$), previously treated patients seemed to have a higher incidence of regional recurrences at level VI (6/10, 60.0%) compared with the other patients (13/48, 27.1%). Furthermore, no differences ($p = 0.241$) were detected in terms of distant recurrences. Similarly, no differences in terms of regional recurrences were detected between patients who underwent (47/704) or not (11/115, $p = 0.356$) ND. At any rate, among those who also received level VI clearance (20/449), the associated risk was higher ($p = 0.055$) likely due to a more advanced pathology.

Survival and disease control according to different patterns of failure

Patients were followed for a minimum period of 2 years and a maximum period of 16.4 years. The mean and the median period were 5.3 and 4.8 years, respectively. Eight patients were lost to follow-up: 3 among recurrent patients and 5 not previously treated. 5-year survival estimates are reported in Table IV. All endpoints were highly impaired by the onset of recurrences ($p < 0.001$, LR and GBW) compared to the non-recurring counterpart. Even if the majority of stratifications did not indicate significant differences among the different types of local recurrence, OS and DSS were more affected by local recurrences of

Table IV. Five-year Kaplan–Meier estimates of assessed oncologic outcomes.

	OS	DSS	DFS	FFL	LFS	LEDFS
Patients with recurrent tumours						
T						
Endolaryngeal	57.8%	69.7%	7.8%	42.5%	33.2%	16.5%
Extralaryngeal	64.8%	88.9%	0.0%	38.1%	27.8%	0.0%
Indeterminate	28.6%	28.6%	0.0%	47.6%	28.6%	14.3%
N						
Level II-V	42.5%	63.9%	5.3%	74.8%	37.1%	25.1%
Level VI	51.2%	63.3%	11.1%	59.5%	41.4%	20.8%
M						
Positive	40.0%	43.6%	9.5%	81.3%	33.9%	21.8%
Patients with non-recurrent tumours	93.4%	98.9%	100%	99.8%	93.2%	93.1%

OS: overall survival; DSS: disease-specific survival; DFS: disease-free survival; FFL: freedom from laryngectomy; LFS: laryngectomy-free survival; LEDFS, laryngo-oesophageal dysfunction-free survival.

undetermined origin. These were, indeed, more prone to induce the exitus of patients with respect to both intralaryngeal ($p < 0.05$, OS, and $p < 0.01$, DSS) and clearly extralaryngeal ($p < 0.05$ OS and DSS) recurrences.
Comparing previously treated and untreated patients, there was no significant differences, with the exception of the incidence of recurrence on regional lymph node ($p < 0.01$). This was likely because previous treatment already involved this site.

Risk analysis on the development of recurrences

At univariate analysis, all variables assessed correlated ($p < 0.05$) with development of recurrences. Nevertheless, at logistic regression, significantly correlated variables were pN classification ($p < 0.001$), presence of cartilage involvement ($p < 0.001$), presence of perineural invasion ($p < 0.05$) and type of surgical treatment (OPHL type III, $p < 0.05$) (Table V).
The onset of recurrences was detected in the 30.6% of patients diagnosed with pN+ cancers and in 10.5% of pN0 (RR = 3.1). Similarly, the frequencies of recurrence were higher in patients with cartilage invasion, both the inner cortex or through the outer cortex, (30.1%) and/or perineural invasions (24.7%), compared to those without these aspects (8.9% RR = 3.4 and 10.4% RR = 2.4, respectively). More extended OPHL type III entailed a 25.4% of recidivate patients, whereas the onset of recurrence after OPHL type II was limited to 11.0% (RR = 2.3). At further analysis, OPHL type III was more frequently employed in patients with previous treatment and in glottic tumours (both $p < 0.001$), though being responsible for a higher incidence of recurrences at level VI ($p < 0.01$).

Discussion

T and N staging are recognised as the best factor to predict survival of patients with laryngeal cancer [20 21 24]. Notwithstanding, prognosis is generally more complex and also depends on many other variables, such as age, clinical-pathologic characteristics, surgical margin status and type of surgery. Some of these could be responsible of adverse events [22], complications or even recurrences that hamper survival of patients [16]. Thus, the literature is rich in manuscripts describing demographic, clinical, pathological and therapeutic variables that have been evaluated as possible prognostic factors for local control of disease and overall survival in patients affected by laryngeal cancer.
On the other hand, evaluation of prognostic factors that are expendable in predicting the onset of recurrences are not so common. The data emerging from this analysis do not correspond exactly to the few studies that have performed appropriate statistical assessment on these factors and the possible inter-relationship between them [23].
Among the few, in the analysis of 253 patients affected glottic and supraglottic laryngeal cancer and undergoing OPHL, Gallo and colleagues [20] demonstrated that the presence of positive surgical margin was the unique significant prognostic factor correlated with the onset of local recurrences. However, we should keep in mind that this was a monocentric study, homogenous regarding indications and surgical techniques and no patients were included with previous treatment for laryngeal cancer. Indeed, with regards to the major differences found between the study by Gallo and coworkers [20], focused on the parameters influencing local control and survival after

Table V. Analyses of factors predicting recurrence.

Variable	Univariate analysis		Logistic regression model	
	Score test	P value	Score test	P value
Previous treatments	0.604	0.437	0.038	0.846
Type of surgery	14.118	< 0.001	4.928	0.026
Tumour site	1.131	0.288	0.932	0.334
pT subcategory	16.389	< 0.001	0.100	0.752
pN ≥ 1 staging	15.742	< 0.001	15.643	< 0.001
Grading	6.481	0.011	1.299	0.254
Cartilage involvement	38.174	< 0.001	24.221	< 0.001
Vascular invasion	3.725	0.054	0.857	0.355
Perineural invasion	15.062	< 0.001	6.018	0.014
Delphian node pN+	5.033	0.025	0.155	0.694
Extranodal extension	16.620	< 0.001	2.253	0.133
Status of margins	6.124	0.013	1.253	0.263
Adjuvant treatments	14.393	< 0.001	0.859	0.354

supracricoid laryngectomy, it should be noted that neither T category nor positivity of resection margins correlated with the risk of recurrence.

The reason may lie in the different types of surgery adopted. In addition, in order to point out the factors that underlie the onset of recurrences (n = 108, 13.2%) in a broader cohort of patients (n = 819), we analysed oncologic results. To obtain this, we subdivided patients accordingly to their pattern of failure. All oncologic outcomes (OS, DSS, DFS, FFL, LFS, and LEDFS) were significantly impaired by the onset of recurrences though the entity was similar among the different pattern on T, N and M. However, local recurrences (T) of indeterminate origin, meaning an iceberg-like recurrence affecting in small part the remnant endolarynx, but massively the paralaryngeal tissues, were more prone to induce exitus (OS and DSS) of patients with respect to the intralaryngeal and extralaryngeal ones. In fact, in these cases it is practically impossible to carry out salvage resection in free and safe margins because diagnosis of recurrence was often late.

Considering that, as expected, recurrent patients had worse prognosis than non-recurrent ones, we evaluated the characteristics of the two populations to detect possible risk factors. Among the 13 different variables assessed, at logistic regression analysis only pN ≥ 1, cartilage invasion (both the inner cortex or through the outer cortex), perineural invasion and employment of more invasive OPHL type III were directly correlated with the onset of recurrences.

N classification has been often related to worse prognosis. In this point of view, it should not be surprising that patients classified as pN≥1 had a higher risk (RR = 3.1) to generally develop recurrences than those without lymph node involvement (pN0). Neck dissection was performed with either elective or curative purposes in the majority of patients (85.9%). Nevertheless, neck recurrences were detected in 25 of 98 (25.5%) pN+ patients, of whom 14 (14.3%) had undergone adjuvant radiotherapy or radiochemotherapy, and in 33 of 721 (4.6%) pN0 patients. Interestingly, recurrences were predominantly at levels II-V in pN+ patients (88.0%), but were equally detected at both level VI (48.5%) and levels II-V (51.5%) in pN0 patients. This different distribution was likely due to the cancer localisation, as recurrence at levels II-V are more frequent in supraglottic tumours. In fact, lymph node relapsing patients were affected by supraglottic cancer in 56.0% of pN+ cases, if compared with the 24.2% of those pN0 ($p < 0.05$). Nevertheless, no differences in terms of tumour classification or use of adjuvant therapy were detected between relapsing and non-relapsing pN+ patients.

In the present cohort, patients with cartilage invasion at the inner cortex or through the outer cortex showed a greater probability to develop recurrences (RR = 3.4). This phenomenon is because the cartilage invasion demonstrates an extralaryngeal escape pathway. This aspect, if suspected pre-operatively, should always determine the recourse to a ND of the ipsilateral levels II-IV, but in particular to meticulous dissection of the central compartment of the neck as well as strap muscles resection and dissection of the pre-laryngeal tissue, immediately analysed by frozen sections. Considering the pattern of failure, the involvement of cartilages had no effect on T and M recurrence, but significantly correlated with that on N. Indeed, cartilage invasion increased the rate of recurrences on level VI lymph nodes (14/50, 28.0%, $p < 0.05$) rather than level II-V ones (10/50, 20.0%, $p < 0.01$), compared to patients with negative cartilage (5/58, 8.6%; and 29/58, 50.0%, respectively). The lack of significance at logistic regression of T classification, despite being strongly related to depth of cartilage invasion seems plausible to us because, beyond T4 tumours, many lesions classified as T3 had focal or multifocal invasion of internal lamina of thyroid cartilage. This phenomenon occurs more frequently in the lower aspect of the thyroid cartilage since the tumour often shows progression towards the crico-thyroid space. Even in the absence of full-thickness cartilage invasion, the behaviour of this subcategory of tumours, schematically represented by a subglottic extension more than 10 mm at the true vocal cord (TVC) midline, is to be considered similar to that of tumours staged in the upper category of T4a. Therefore, more cautious surgical resection should be preferred, characterised by careful dissection of level VI nodes.

For what concerns perineural invasion, although enhancing the overall risk of developing recurrences (RR = 2.4), the risk factor did not correlate *per se* with significant alteration of the pattern of failure. However, it is generally present in tumours characterised by advanced T stage and high histological grade classification, which are thus more refractory to treatments and show worse prognosis. Patients with perineural invasion should undergo strict follow-up to diagnose recurrences early. Moreover, this result gives rise to discussions in the tumour board about the necessity to treat patients affected by locally advanced tumours with adjuvant radiotherapy, possibly showing this negative prognostic factor for loco-regional recurrence.

Finally, regarding the type of surgery, we detected a higher risk of developing recurrences in cases treated by OPHL type III (RR = 2.3). We should keep in mind that supratracheal laryngectomies (OPHL type III) are generally employed in the treatment of more aggressive and advanced disease. Thus, more attention should be paid to this analysis.

Undoubtedly, OPHL type III extended indications of partial laryngectomy in case of cancers (glottic-subglottic tumours, T4a tumours) that, historically, were treated by total laryngectomy. Nevertheless, it determines a major rate of close margins, negative the frozen sections when the sample is collected on the side of the remnant larynx, but positive on the side of the specimen, generally correlates with a major rate of level VI lymph nodes metastasis. Although not mathematically, considering that the larynx also "accepts" minimal resection margins, this can expose patients to a greater number of loco-regional relapses (compared to those after OPHL type II) especially when detected as T indeterminate feature recurrences or on the level VI lymph nodes. Therefore, cases to be addressed to this surgical option must be selected very well.

OPHLs type III are safely indicated in case of T3 tumours with subglottic extension preferable < 10 mm at the TVC midline or in case of T4a "anterior" tumour [15], because these patients have demonstrated good oncologic results with a low percentage of recurrence. On the contrary, even T3 or T4a "posterior" tumours, where arytenoid fixity is related to an infraglottic extension > 10 mm at the TVC midline or to a massive posterior crico-arytenoid unit invasion, the tumour clearly acts aggressively as if it were always a T4.

OPHLs type III surgeries are therefore quite safe in first two cases, especially if the resection encompasses one crico-arytenoid unit. In the latter conditions, the safest treatment is total laryngectomy, also considering some disappointing events (positive margins, level VI pN+, ENE), that are more frequently associated with recurrence. Only in the case of categorical refusal by the patient to total laryngectomy, after informing about the risk of giving priority to the functional outcomes rather than to oncologic ones, in some very selected cases it is still possible to carry out an OPHL type IIIa. This option, even if not optimal, should be equally considered as chemoradiotherapy when drafting guidelines.

Conclusions

To our knowledge, this study is the first to use logistic regression on a very large cohort of patients treated by all different types of supracricoid and supratracheal partial laryngectomies (OPHL type II / III). The detection of clinical and pathological parameters that correlate with the development of recurrences (pN classification, cartilage involvement, perineural invasion, and thus the type of surgical treatment adopted) can be useful to reduce the event rate. Therefore, we suggest serious discussion in the multidisciplinary tumour board regarding the possible indications of OPHL, rather than resorting to a simpler and safer total laryngectomy or, alternatively, to concomitant chemoradiation-therapy, especially in cases where additional risk factors are present, such as the exiguity of surgical margins and use of more extensive surgeries.

In this delicate process, the patient and his/her caregivers also are involved, being important actors. With due delicacy, also analysing data from large series, they must be faced with and helped in choosing among the difficult dichotomy to give greater emphasis to a "*quoad vitam*" rather than to a "*quoad functionem*" prognosis.

Acknowledgements

This paper was awarded with the first prize of the Italian Society of Otorhinolaryngology, Head and Neck Surgery, Caliceti - Citelli - Pietrantoni at the 105th SIOeChCF National Meeting, Napoli May 16-19, 2018. These researches have been supported by "FPRC 5xmille Ministero Salute 2016 (Progetto "Approccio di ricerca diagnostico e terapeutico al paziente con Tumori del Colon Retto del distretto cervico-facciale ARIDTE-Biohenec).

Conflict of interest statement

None declared.

References

1 Succo G, Bussi M, Presutti L, et al. *Supratracheal laryngectomy: current indications and contraindications*. Acta Otorhinolaryngol Ital 2015;35:146-56.

2 Bernier J, Domenge C, Ozsahin M, et al. *European Organization for Research and Treatment of Cancer Trial 22931. Postoperative irradiation with or without concomitant chemotherapy for locally advanced head and neck cancer.* N Engl J Med 2004;350:1945-52. https://doi.org/10.1056/NEJMoa032641.

3 Succo G, Crosetti E, Bertolin A, et al. *Benefits and drawbacks of open partial horizontal laryngectomies, Part A: early- to intermediate-stage glottic carcinoma*. Head Neck 2016;38(Suppl 1):E333-40. https://doi.org/10.1002/hed.23997.

4 Peretti G, Piazza C, Mensi M, et al. *Endoscopic treatment of cT2 glottic carcinoma: prognostic impact of different pT subcategories*. Ann Otol Rhinol Laryngol 2005;114:579-86. https://doi.org/10.1177/00034894051140080.

5 Succo G, Crosetti E, Bertolin A, et al. *Benefits and drawbacks of open partial horizontal laryngectomies, Part B: intermediate and selected advanced stage laryngeal carcinoma*. Head Neck 2016;38 (Suppl.1):E649-57. https://doi.org/10.1002/hed.24064.

6 Tomeh C, Holsinger FC. *Laryngeal cancer*. Curr Opin Otolaryngol Head Neck Surg 2014;22:147-53. https://doi.org/10.1097/MOO.0000000000000032.

7 Forastiere AA, Zhang Q, Weber RS, et al. *Long-term results of RTOG 91-11: a comparison of three nonsurgical treatment strategies to preserve the larynx in patients with locally advanced larynx cancer*. J Clin Oncol 2013;31:845-52. https://doi.org/10.1200/JCO.2012.43.6097.

[8] Olsen K. *Reexamining the treatment of advanced laryngeal cancer*. Head Neck 2010:32:1-7. https://doi.org/10.1002/hed.21294.

[9] Benito J, Holsinger FC, Perez-Martin A, et al. *Aspiration after supracricoid partial laryngectomy: incidence, risk factors, management, and outcomes*. Head Neck 2011;33:679-85. https://doi.org/10.1002/hed.21521.

[10] Sobin LH, Gospodarowicz MK, Wittekind C. *TNM classification of malignant tumours*. 7th edition. Hoboken, NJ: Wiley-Blackwell; 2011.

[11] Karnofsky D, Burchenal J. *The clinical evaluation of chemotherapeutic agents in cancer*. In: MacLeod CM, editor. *Evaluation of Chemotherapeutic agents.* New York: Columbia University Press; 1949. pp. 191-205.

[12] Pinar E, Imre A, Calli C, et al. *Supracricoid partial laryngectomy: analyses of oncologic and functional outcomes*. Otolaryngol Head Neck Surg 2012:147:1093-8. https://doi.org/10.1177/0194599812457334.

[13] Rizzotto G, Crosetti E, Lucioni M, et al. *Oncological outcomes of supratracheal laryngectomy: a critical analysis*. Head Neck 2015;37:1417-24. https://doi.org/10.1002/hed.23773.

[14] Rizzotto G, Crosetti E, Lucioni M, et al. *Subtotal laryngectomy: outcomes of 469 patients and proposal of a comprehensive and simplified classification of surgical procedures*. Eur Arch Otorhinolaryngol 2012;269:1635-46. https://doi.org/10.1007/s00405-012-1928-4.

[15] Succo G, Crosetti E, Bertolin A, et al. *Treatment of T3-T4a laryngeal cancer by open partial horizontal laryngectomies: prognostic impact of different pT subcategories*. Head Neck 2018;40:1897-908. https://doi.org/10.1002/hed.25176.

[16] Crosetti E, Caracciolo A, Molteni G, et al. *Unravelling the risk factors that underlie laryngeal surgery in elderly*. Acta Otorhinolaryngol Ital 2016:36:185-93. https://doi.org/10.14639/0392-100X-817.

[17] Succo G, Peretti G, Piazza C, et al. *Open partial horizontal laryngectomies: a proposal for classification by the working committee on nomenclature of the European Laryngological Society*. Eur Arch Otorhinolaryngol 2014;271:2489-96. https://doi.org/10.1007/s00405-014-3024-4.

[18] Robbins KT, Medina JE, Wolfe GT, et al. *Standardizing neck dissection terminology. Official report of the Academy's Committee for Head and Neck Surgery and Oncology.* Arch Otolaryngol Head Neck Surg 1991;117:601-5.

[19] Ang K. *Larynx preservation clinical trial design: summary of key recommendations of a consensus panel*. Oncologist 2010;15(Suppl 3):25-9. https://doi.org/10.1634/theoncologist.2010-S3-25.

[20] Gallo A, Manciocco V, Simonelli M, et al. *Supracricoid partial laryngectomy in the treatment of laryngeal cancer: univariate and multivariate analysis of prognostic factors*. Arch Otolaryngol Head Neck Surg 2005:131:620-5. https://doi.org/10.1001/archotol.131.7.620.

[21] LeBlanc B, Shi R, Mehta V, et al. *Improvements in survival and disparities for advanced-stage laryngeal cancer*. JAMA Otolaryngol Head Neck Surg 2015:141:169-73. https://doi.org/10.1001/jamaoto.2014.2998.

[22] Pezier T, Nixon I, Joshi A, et al. *Pre-operative tracheostomy does not impact on stomal recurrence and overall survival in patients undergoing primary laryngectomy*. Eur Arch Otorhinolaryngol 2013;270:1729-35. https://doi.org/10.1007/s00405-012-2213-2.

[23] Joo YH, Park JO, Cho KJ KM. *Relationship between paraglottic space invasion and cervical lymph node metastasis in patients undergoing supracricoid partial laryngectomy*. Head Neck 2012:34:1119-22. https://doi.org/10.1002/hed.21892.

[24] Atallah I, Berta E, Coffre A, et al. *Supracricoid partial laryngectomy with crico-hyoido-epiglottopexy for glottic carcinoma with anterior commissure involvement*. Acta Otorhinolaryngol Ital 2017;37:188-94. https://doi.org/10.14639/0392-100X-1002.

Received: November 1, 2018 - Accepted: January 11, 2019

How to cite this article: Crosetti E, Bertolin A, Molteni G, et al. *Patterns of recurrence after open partial horizontal laryngectomy types II and III: univariate and logistic regression analysis of risk factors.* Acta Otorhinolaryngol Ital 2019;39:235-243. https://doi.org/10.14639/0392-100X-2409

Address for correspondence: Erika Crosetti, Head and Neck Oncology Service, FPO IRCCS, Candiolo Cancer Institute, Candiolo (TO), Italy. Tel. +39 011 9933663. E-mail: erika.crosetti@ircc.it

ACTA OTORHINOLARYNGOLOGICA ITALICA 2019;39:244-249; doi: 10.14639/0392-100X-N0221

Laryngology

Time as a factor during endoscopic assessment of swallowing: relevance in defining the score and severity of swallowing disorders

Il tempo come fattore durante la valutazione endoscopica della deglutizione: la sua rilevanza nel definire il punteggio e la gravità dei disturbi della deglutizione

D. FARNETI[1], B. FATTORI[2], L. BASTIANI[3]

[1] Audiology and Phoniatrics Department, Romagna Health Service, Rimini Hospital, Italy; [2] ENT Department, Audiology and Phoniatrics, Pisa University, Italy; [3] Institute of Clinical Physiology, National Council of Research (CNR), Pisa, Italy

SUMMARY

Time is a parameter of great interest in swallowing and can be considered in different ways to express severity during endoscopic evaluation. The objectives of this study are to evaluate how the severity of this score changes at different times of scoring and the interaction between residue persistence and airway invasion. Two experienced raters blindly evaluated 35 short clips of bolus transit that were recorded during endoscopic evaluations of 16 patients with dysphagia of differing aetiologies. The pooling score (p-score) and the Penetration Aspiration Scale (PAS) were detected after the first swallowing (T1) and after the fifth dry swallow (T5). For each task, the time needed to complete the clearing of the bolus (total time: TT) was blindly determined by the two raters and compared with the Functional Oral Intake Scale (FOIS) and Dysphagia Outcome and Severity Scale (DOSS) scales, previously detected.The inter-rater agreement between the 2 raters in scoring the p-score and PAS was good (ICC > 0.800) for T1 vs T1 and T1 vs T5, and in determining TT for each consistency (ICC > 0.9), with a Spearman's Rho > 0.70 and > 0.90 respectively. A statistical correlation of the p-score total, TT and consistency with FOIS and DOSS was found. The p-score showed a good trade-off between sensitivity and specificity compared with the PAS aspiration and penetration scores. The time of scoring (T1 vs T5) is relevant in detecting severity of dysphagia during endoscopic evaluation. The time spent to clear residue is a useful parameter and is correlated with severity of dysphagia expressed by the p-score and with functional swallowing status in dysphagic patients. The p-score is correlated with the PAS score in detecting airway invasion.

KEY WORDS: Swallowing • Deglutition disorders • Endoscopy • Aspiration • Residue • Severity

RIASSUNTO

Il tempo è un parametro di grande interesse nella deglutizione variamente considerato per esprimere gravità durante una valutazione endoscopica. Gli obiettivi del lavoro sono di valutare come la gravità di un punteggio cambia in diversi tempi di rilevazione e l'interazione tra persistenza di residui e invasione delle vie aeree. Due valutatori esperti hanno valutato in cieco 35 brevi filmati di transiti faringei di bolo, registrati durante valutazioni endoscopiche di 16 pazienti con disfagia a differente eziologia. Il punteggio del pooling score (p-score) e la Penetration Aspiration Scale (PAS) sono stati rilevati dopo la prima deglutizione (T1) e dopo la quinta (T5). Per ogni transito, il tempo necessario per completare la detersione del bolo (tempo totale: TT) è stato determinato in cieco dai due valutatori e confrontato con le scale Functional Oral Intake Scale (FOIS) and Dysphagia Outcome and Severity Scale (DOSS) scales, precedentemente determinate. L'affidabilità inter-rater tra i 2 valutatori nel punteggio del p-score e PAS è stata buona (ICC > 0,800) per T1 vs T1 e T1 vs T5, e nel determinare TT per ogni consistenza (ICC > 0,9), con Rho di Spearman > 0,70 e > 0,90 rispettivamente. È stata trovata una correlazione statistica fra il p-score totale, TT, e la consistenza con il FOIS e il DOSS. Il p-score ha mostrato un buon trade-off tra sensibilità e specificità rispetto ai punteggi PAS che esprimono aspirazione e penetrazione. Il tempo di applicazione di una scala (T1 vs T5) è rilevante nel determinare la severità della disfagia durante una valutazione endoscopica. Il tempo impiegato dal paziente per detergere i residui è un parametro utile da determinarsi ed è correlato alla gravità della disfagia espressa dal p-score e allo stato funzionale di pazienti disfagici. Il p-score è correlato al punteggio PAS nel rilevare l'invasione delle vie aeree.

PAROLE CHIAVE: *Deglutizione • Disturbi della deglutizione • Endoscopia • Aspirazione • Ristagni • Gravità*

Introduction

In normal conditions, the progression of the bolus through the upper digestive tract must meet criteria of efficiency and effectiveness: any condition that deviates from these criteria generates an unsafe swallowing act with nasal regurgitation, penetration, aspiration, or an inefficient swallowing act, with residue of bolus [1]. These two conditions can coexist and influence each other: a bolus that leaves residue in the pharynx can be cleared or can invade the

upper airway during a subsequent swallowing act [2], while a bolus invading the upper airway can indefinitely remain in the larynx or cervical trachea. Instrumental investigation must be able to document these conditions, with the aim of defining criteria of severity that better indicate, in a clinical context, the risk of respiratory or nutritional complications [3 4]. This same information, in the short term, is useful in planning therapeutic strategies and in reducing long-term complications [5]. Today, several tools are available to evaluate and quantify ineffective or inefficient swallowing. Some tools designed for the radiological setting have been adapted and applied to endoscopic examination. An example is the penetration aspiration scale (PAS) [6], originally designed for the modified barium swallow (MBS) and subsequently replicated for endoscopy [7-10]. The PAS is widely used to define the entity of airway invasion and possible ejection attempts. The PAS score is applied after the first/second swallow, without a standard reported in the literature or in the original paper [6]. However, in clinical practice, more varied conditions can be found. A bolus can be propelled by more swallowing acts (multiple swallowing) or coexist with residue, imposing several dry swallows before clearing. In these conditions, airway invasion may occur after the swallow [11] or require defensive strategies (spontaneous or requested) carried out over a longer period of time (more than one or two dry swallows).

In the literature, some rating scales are applied after the first-second swallowing acts, such as the aforementioned PAS, and others after the second one, such as the pooling score (p-score) [12] and Boston Residue and Clearance Scale [13]. Some scales do not mention the time of scoring [8] and others leave the decision to the clinician as to when to score [14-18]. The first tool proposed, to be applied after the second swallow, was the p-score, a validated scale [12 19] that considers 5 dry swallows before scoring residue in the pharyngeal or laryngeal cavities. The total score derives from the sum of partial scores attributed to the anatomical site where material is pooling, its amount (semi-quantitatively compared to the volume's bidimensional viewing of the cavities) and to the number and effectiveness of dry swallowing, or other strategies, performed in the attempt to clear residue ("management" of residue, see Table I). The score expresses a continuum of severity which, in clinical practice, can be spread over four levels, describing different levels of clinical severity (Table I). The consistency of the bolus does not seem to affect the score [19]. The p-score, like the other scores, does not consider the parameter 'time', i.e. the time necessary to clear the bolus or to complete the 5 dry swallows requested, but focuses on the residue and the correlates that define it.

Table I. Pooling score (p-score).

Pooling	Endoscopic landmark	
Site	Vallecule – marginal zone	1
	Pyriform sinus	2
	Vestibule – vocal cords	3
	Below the vocal cords	4
Amount	Coating	1
	Minimum	2
	Maximum	3
Management	< 2	2
	2 > < 5	3
	> 5	4
Score	P 4-11	

"Pooling" any material that dwells or coats the hypopharynx and/or larynx cavities before/after swallowing. Site: anatomical landmark in a cranio-caudal direction. Amount: volumetric ratio between content and container (minimally filled, less than half or more than half). Management: any spontaneous/reflex activity adopted to clear pooling (dry swallows, gurgling, clearing and cough). p- score: 4-5 = minimum score, corresponding to no endoscopic signs of dysphagia; 6-7 = low score, corresponding to mild dysphagia; 8-9 = middle score, corresponding to moderate dysphagia; 10-11 = high score, corresponding to severe dysphagia

Swallowing times during endoscopic evaluation of swallowing have been assessed in several studies [20-25] and time association with swallowing abnormalities was explored in some of them [22-25]. Nevertheless, the time considered in those investigations does not include the time needed to clear residue, a parameter which has previously been considered in our preliminary report [26]. Cinical observations, indeed, show conditions where the material pooling is cleared in a few seconds and other conditions where a longer time is needed to match this goal, provided that it is possible. In this context, the time of 'management' of the residue recalls the attempts to clear saliva with spontaneous swallowing acts performed by elderly patients [15] or stroke patients [27], although different pressure and awareness are required to clear residue. This perspective leads to a parameter of severity where the clearing time can become a marker of effectiveness of the swallowing act in pathological conditions or where its increase, under stress, can become a marker of fatigue [28 29]. If this were true, the time of 'management' could vary similarly to the variation of other scales [26] such as the Functional Oral Intake Scale (FOIS) [30] and the Dysphagia Outcome and Severity Scale (DOSS) [31].

With these considerations in mind, the aims of this study are to evaluate: 1) if the p-score changes when applied before the fifth swallowing act and whether the PAS changes when applied after the first; 2) the reliability of 2 raters in scoring residue at different times and their reliability in determining these times (time of 'management' of the p-score); 3) possible correlation of PAS to p-score.

Materials and methods

In a prospective way, 16 consecutive outpatients (11M/5F, mean age 63.94 years ± 15.46, range 25-88) were submitted to a fibreoptic endoscopic evaluation of swallowing (FEES) [32]. The patients were complaining of swallowing disorders due to different aetiologies (Table II). Inclusion criteria were: over 18 years old, an instrumentally documented impaired swallow (residue, false routes), compliance to the endoscopic procedure; exclusion criteria were: less than 18 years old and non-compliance to the endoscopic procedure. The patients with low dysphagia were considered because they respected the criteria of inclusion. FEES was performed with a Storz endoscope (model 11101RP2, 30 cm long, 3.5 mm in diameter) and recorded with a workstation (Xion medical products GmbH, Berlin Buchholz). During FEES and with the endoscope in place, one bolus of each consistency was given to each patient: 5 cc pureed (P), ¼ of a cracker (regular-R) and 5 cc liquid (L) [33]. The patients prepared the bolus and swallowed without any command. Some patients were not able to test all three consistencies, owing to the severity of their complaint. For each patient, short videos were obtained for each swallowing trial so that a total of 35 clips were collected and reviewed by two expert raters (with more than 15 years' experience in performing FEES) in a blind manner. The raters were requested to score each bolus trial with the p-score and the PAS. Both the p-score and the PAS score were applied after the first (time 1 – T1) and fifth swallow (time 5 – T5). In this way, the parameter 'management' of the p-score was always the minimum provided by the score. The raters also blindly determined the time necessary to perform the 5 dry swallowing acts (total time: TT). TT was timed with a stopwatch in iOS 9.0, 4+ (Tim O's Studios, LLC) at the beginning of the first white-out and at the conclusion of the fifth white-out [34]. In accordance with the p-score, spontaneous and cued dry swallows were considered. TT was compared with the patients' ability for oral intake of food and liquid, measured against the Functional Oral Intake Scale (FOIS), even if only validated for stroke patients [30] and the functional severity of dysphagia measured against the Dysphagia Outcome and Severity Scale (DOSS) [31]. The scales were previously determined by rater 1. Because of the small sample, monovariate analysis was previously performed among TT and consistencies (explanatory variables) and FOIS and DOSS, respectively. Subsequently, multiple linear regression, considering FOIS and DOSS as dependent variables, was performed taking into account the TT, p-score total and consistency.

Table II. Case series.

Pts n.	Main pathology	Gender	Age
1	Arnol-Chiari malformation	M	56
2	MSA-P	M	85
3	Myasthenia gravis	M	73
4	Vascular dementia	M	74
5	Parkinson's disease	M	75
6	TBI sequelae	M	44
7	Oesophageal dysphagia	F	72
8	Supraglottic laryngectomy	F	80
9	Stroke sequelae	M	88
10	Parkinson's and ictus	M	81
11	Cervical hyperostosis	M	84
12	Steinert syndrome	M	69
13	Klinefelter syndrome	M	25
14	Cerebral palsy	F	26
15	Subtotal laryngectomy	F	58
16	Multiple sclerosis	F	33

The intra-class correlation coefficient was performed to evaluate the inter-rater reliability of the two raters for FEES at T1 and T5 (ICC) and determine TT. In accordance with the literature, the following were considered for ICC values: 0-0.2 poor; 0.3-0.4 fair; 0.5-0.6 moderate; 0.7-0.8 strong; and > 0.8 almost perfect. For each rater, the Rho Spearman's coefficient ($r > 0.70$-sing < 0.05) was performed to evaluate the correlation between PAS and p-score and TT. Furthermore, in order to determine optimal thresholds for the p-score when compared to PAS diagnoses, the Receiver Operator Characteristic (ROC) curve analysis was performed after dichotomising PAS between penetration scores 2 to 5 and aspiration scores 6 to 8. To determine the best balance between sensitivity and specificity, the Youden Index (Y = sensitivity+specificity−1), was chosen as the criterion for cut-off value selection. All statistical analyses were performed using SPSS v.21.0 (IBM Corp., Armonk, NY, USA) and STATA version 13 (STATA Corp., TX, USA).

All patients gave their written consent to the procedures, in accordance with the Declaration of Helsinki. The study was approved by the local Ethical Research Committee.

Results

The mean times necessary to clear residue for P, R and L were 22.5 secs (range 4-42), 30.7 secs (range 11-44) and 16.6 secs (range 8-33), respectively, with a mean TT of 53.8 sec. The inter-rater agreement between the 2 raters in scoring the p-score and PAS was good (ICC > 0.800) for each consistency and time of scoring (T1 vs T1 and

T1 vs T5) with the exception of the liquid bolus. The inter-rater agreement between the 2 raters in detecting TT was good (ICC > 0.9) for each consistency (Spearman's Rho > 0.90-sing < 0.001). A correlation between the PAS and the p-score at T1 vs T1 and T1 vs T5 was observed only for the pureed consistency for rater 1 and for pureed and liquid ones for rater 2 (Spearman's Rho > 0.70-sing < 0.05). The linear regression model documented a significant correlation of the p-score total, TT and consistency with FOIS and DOSS. In particular, increasing the time spent in clearing residue corresponded to an increase in the p-score and decreased the FOIS for all consistencies; increasing the p-score decreased the DOSS score for R and L (Table III, IV). A good correlation between PAS score and p-score was found (Spearman's rho 0.924-P < 0.05). The screening properties of the p-score when compared to the PAS cut-off diagnosis of penetration (scores 2 to 5) and aspiration (scores 6 to 8) showed a good trade-off between sensitivity and specificity compared with the PAS aspiration scores (area under the ROC curve = 0,958; 95% CI = 0.784-0.994) and with the PAS penetration scores (area under the ROC curve = 0.622; 95% CI = 0.352-0.792), with a p-score cut-off of 3 for penetration and of 4 for aspiration, respectively.

Discussion

Our experience shows that when applying the p-score at T1 and the PAS at T5, with FEES, they correlate only for the P consistency for rater 1 and P and L for rater 2. This leads us to consider that the lack of correlation between T1 and T5 suggests a real different value of the score applied, i.e. applying the PAS over the first swallowing act changes the score itself. It is also worth mentioning that for the liquid bolus there was no concordance between the two raters, contrary to the other consistencies.

The parameter 'time' also shows its importance under the quantitative perspective, as the time spent in completing a sequence of dry swallows. In our sample, the mean time necessary to clear residue for P, R and L was far longer than the time physiologically reported in the literature for clearing boluses of the same consistency [20-25]. The detection of this parameter, in our experience, seems to be a reliable parameter worth including in endoscopic evaluation of swallowing [26]. The p-score, which indirectly considers this parameter (residue 'management' in TT) enriched in that sense, may express a further criterion of severity (see Appendix). In our sample, the increase in TT is related to the increase in the p-score, and both are related to the decrease in the FOIS score for all consistencies tested and to the increase of the DOSS scale for R and L [26].

Even the consistency, which does not affect the p-score [19], when related to the TT, seems to be a parameter able to influence the outcome of the swallowing act (presence of residue) performed spontaneously or upon request by the patients in our sample. Increasing TT, a reduction in the efficiency of the swallowing act could be hypothesised: the fate of the residue during the TT is not predictable,

Table III. Linear regression models: relationship between TT and consistencies (explanatory variables) and FOIS score (dependent variable).

	Beta	95% confidence interval		P value
Fees - T - P tot	-1.321	-4.649	-0.635	0.029
Fees - T - P sec	1.128	-0.022	1.216	0.053
Fees - T - R tot	-1.927	-0.801	-1.494	0.004
Fees - T - R sec	-2.437	-0.370	-0.171	0.007
Fees - T - L tot	-0.439	-0.505	-0.080	0.027
Fees - T - L sec	-0.903	-0.113	-0.050	0.007

T = time, P = pureed, R = regular, L = liquid

Table IV. Linear regression models: relationship between TT and consistencies (explanatory variables) and DOSS score (dependent variable).

	Beta	95% confidence interval		P value
Fees - T - P tot	-,294	-,519	,208	,208
Fees - T - P sec	,025	-,034	,039	,826
Fees - T - R tot	-1,338	-,378	-1,215	,015
Fees - T - R sec	-,931	-,173	-,033	,024
Fees - T - L tot	-,488	-,697	-,047	,046
Fees - T - L sec	-,634	-,101	-,014	,030

T = time; P = pureed; R = regular; L = liquid

but it is plausible that it may be related to the aetiology/comorbidities [35 36]. In this sense, a possible correlation between the p-score compared with PAS, in detecting penetration and aspiration, is expressed by the area under the ROC curves: these values indicate a good predictability of the p-score for the three consistencies in terms of sensitivity and specificity. The cut-off for aspiration is 4 and the cut-off for penetration is 3, coinciding with the subparameter 'site' of the p-score, identifying residues below and above the vocal cords, respectively.

The main limitations of this work are the small sample and the different numerical representations of the bolus swallowed in different consistencies. Bearing this in mind, the work is intended to have a preliminary character, and to test the value of the 'time' parameter in defining the clinical severity of a swallowing disorder. Further research is in progress to correlate the tp-score with the fatigability of patients with swallowing disorders due to specific aetiologies.

Conclusions

The parameter 'time' was evaluated applying scores that consider directly (p-score) or indirectly (PAS) the bolus and its fate after subsequent swallowing acts (multiple swallows or cued swallows). The evaluation of FEES clips of swallowing tasks suggests how, by applying the p-score at T1 and PAS at T5, although for different consistencies, produced different scores between two expert raters. We conclude that the time of detection of a score modifies the score, so that the time of scoring (the first or subsequent swallowing act) should be previously defined and considered.

The time needed by the patient to clear the residue is a reliable parameter that correlates with the severity of the p-score and other scales that relate to the patients' functional status or with their deglutition skills, suggesting the possibility of a clinical use of the tp-score (Appendix) in the follow-up of patients with swallowing disorders due to specific aetiologies or after stressful swallowing activities (fatigue detection).

Conflict of interest statement

None declared.

References

1 Clavé P, Verdaguer A, Arreola V. *Oropharyngeal dysphagia in the elderly*. Med Clin (Barc) 2005;124:742-8.

2 Farneti D, Consolmagno P. *Aspiration: the predictive value of some clinical and endoscopic signs. Evaluation of our case series.* Acta Otorynolaryngol Ital 2005;25:36-42.

3 Farneti D, Genovese E. *Endoscopic criteria in assessing severity of swallowing disorders*. In: Speyer R, Bogaardt H, editors. *Seminars in Dysphagi*a. London,UK: InTech; 2015. pp. 71-90.

4 Farneti D. *The instrumental gold standard: FEES*. J Gastroenterol Hepatol Res 2014;3:1281-91.

5 Brady S. *Use of dysphagia severity scales during fiberoptic endoscopic exam of swallowing: treatment decisions and planning*. ASHA Special Interest Division 13, Perspectives in Swallowing and Swallowing Disorders 2007;16:10-3.

6 Rosenbek JC, Robbins, J Roecker EV, et al. *A Penetration-aspiration scale*. Dysphagia 1996;11:93-8.

7 Colodny N. *Interjudge and intrajudge reliabilities in Fiberoptic Endoscopic Evaluation of Swallowing (Fees®) using the Penetration–Aspiration Scale: a replication study*. Dysphagia 2002;17:308-15. https://doi.org/10.1007/s00455-002-0073-4.

8 Kelly AM, Leslie P, Beale T, et al. *Fibreoptic endoscopic evaluation of swallowing and videofluoroscopy: does examination type influence perception of pharyngeal residue severity?* Clin Otolaryngol 2006;31:425-32. https://doi.org/10.1111/j.1749-4486.2006.01292.x.

9 Kelly AM, Drinnan MJ, Leslie P. *Assessing penetration and aspiration: how do videofluoroscopy and fiberoptic endoscopic evaluation of swallowing compare?* Laryngoscope 2007;11:1723-7. https://doi.org/10.1097/MLG.0b013e318123ee6a.

10 Pilz W, Vanbelle S, Kremer B, et al. *Observers' agreement on measurements in fiberoptic endoscopic evaluation of swallowing*. Dysphagia 2016;31:180-7. https://doi.org/10.1007/s00455-015-9673-7.

11 Molfenter SM, Steele CM. *The relationship between residue and aspiration on the subsequent swallow: an application of the normalized residue ratio scale*. Dysphagia 2013;28:494-500. https://doi.org/10.1007/s00455-013-9459-8.

12 Farneti D. *Pooling score: an endoscopic model for evaluating severity of dysphagia*. Acta Otorhinolaryngol Ital 2008;28:135-40.

13 Kaneoka AS, Langmore SE, Krisciunas GP, et al. *The Boston Residue and Clearance Scale: preliminary reliability and validity testing*. Folia Phoniatr Logop 2013;65:312-7. https://doi.org/10.1159/000365006.

14 Neubeuer PD, Rademake AW, Leder S. *The Yale Pharyngeal Residue Severity Rating Scale: an anatomically defined and image-based tool*. Dysphagia 2015;30:521-8. https://doi.org/10.1007/s00455-015-9631-4.

15 Murray J, Langmore SE, Ginsberg S, et al. *The significance of accumulated oropharyngeal secretions and swallowing frequency in predicting aspiration*. Dysphagia 1996;11:99-103.

16 Park WY, Lee TH, Ham NS, et al. *Endoscopist-directed flexible endoscopic evaluation of swallowing to the videofluoroscopic swallowing study increased the detection rates of penetration, aspiration, and pharyngeal residue*. Gut Liver 2015;9:623-8. https://doi.org/10.5009/gnl14147.

17 Tohara H, Nakane A, Murata S, et al. *Inter- and intra-rater reliability in fibroptic endoscopic evaluation of swallowing*. J Oral Rehabil 2010;37:884-91. https://doi.org/10.1111/j.1365-2842.2010.02116.x.

18 Pilz W, Baijens LW, Passos VL et al. *Swallowing assessment in myotonic dystrophy type 1 using fiberoptic endoscopic evaluation of swallowing (FEES)*. Neuromuscul Disord 2014;24:1054-62. https://doi.org/10.1016/j.nmd.2014.06.002.

19 Farneti D, Fattori B, Nacci A, et al. *The Pooling-score (P-score): inter- and intra-rater reliability in endoscopic assessment of the severity of dysphagia*. Acta Otorhinolaryngol Ital 2014;3:105-10.

20 Dua KS, Ren J, Bardan E, et al. *Coordination of deglutitive glottal function and pharyngeal bolus transit during normal eating*. Gastroenterology 1997;112:73-83.

[21] Butler SG, Stuart A, Kemp S. *Flexible endoscopic evaluation of swallowing in healthy young and older adults*. Ann Otol Rhinol Laryngol 2009;118:99-106. https://doi.org/10.1177/000348940911800204.

[22] Butler SG, Maslan J, Stuart A, et al. *Factors influencing bolus dwell times in healthy older adults assessed endoscopically*. Laryngoscope 2011;121:2526-34. https://doi.org/10.1002/lary.22372.

[23] Tejima C, Kikutani T, Takahashi N, et al. *Application of simple swallowing provocation test with fiberoptic endoscopic evaluation of swallowing in a cross-sectional study*. BMC Geriatrics 2015;15:48. https://doi.org/10.1186/s12877-015-0049-5.

[24] Ursino S, Seccia V, Cocuzza P, et al. *How does radiotherapy impact swallowing function in nasopharynx and oropharynx cancer? Short-term results of a prospective study*. Acta Otorhinolaryngol Ital 2016;36:174-184. https://doi.org/10.14639/0392-100X-640.

[25] Wan P, Chen X, Zhu L, et al. *Dysphagia post subcortical and supratentorial stroke*. J Stroke Cerebrovasc Dis 2016;25:74-82. https://doi.org/10.1016/j.jstrokecerebrovasdis.2015.08.037.

[26] Farneti D, Fattori B, Bastiani L. *Residue and clearing time: preliminary considerations in defining dysphagia severity in daily practice*. Hearing, Balance and Communication 2019. https://doi.org/10.1080/21695717.2019.1631047.

[27] Crary MA, Carnaby GD, Sia I, et al. *Spontaneous swallowing frequency has potential to identify dysphagia in acute stroke*. Stroke 2013;44:3452-7. https://doi.org/10.1161/STROKEAHA.113.003048.

[28] Miller N, Noble E , Jones D, et al. *Hard to swallow: dysphagia in Parkinson's disease*. Age Ageing 2006;35:614-18. https://doi.org/10.1093/ageing/afl105.

[29] Solomon NP. *What is orofacial fatigue and how does it affect function for swallowing and speech?* Semin Speech Lang 2006;27:268-82. https://doi.org/10.1055/s-2006-955117.

[30] Crary MA, Carnaby-Mann GD, Groher ME, et al. *Initial psychometric assessment of a functional oral intake scale for dysphagia in stroke patients*. Arch Phys Med Rehabil 2005;86:1516-20. https://doi.org/10.1016/j.apmr.2004.11.049.

[31] O'Neil KH, MA, Purdy M, Falk J, et al. *The Dysphagia Outcome and Severity Scale*. Dysphagia 1999;14:139-45. https://doi.org/10.1007/PL00009595.

[32] Langmore SE, Schatz K, Olson N. *Endoscopic and videofluoroscopic evaluations of swallowing and aspiration*. Ann Otol Rhinol Laryngol 1991;100:678-81. https://doi.org/10.1177/000348949110000815.

[33] Cichero JA, Steele C, Duivestein J et al. *The need for international terminology and definitions for texture-modified foods and thickened liquids used in dysphagia management: foundations of a global initiative*. Curr Phys Med Rehabil Rep 2013;24:280-91. https://doi.org/10.1007/s40141-013-0024-z.

[34] Mozzanica F, Lorusso R, Robotti C, et al. *Effect of age, sex, bolus volume, and bolus consistency on whiteout duration in healthy subjects during FEES*. Dysphagia 2019;34:192-200. https://doi.org/10.1007/s00455-018-9961-0.

[35] Farneti D, Turroni V, Genovese E. *Aspiration: diagnostic contributions from bedside swallowing evaluation and endoscopy*. Acta Otorhinolaryngol Ital 2018;38:511-6. https://doi.org/10.14639/0392-100X-1967.

[36] Nordio S, Di Stadio A, Koch I, et al. *The correlation between pharyngeal residue, penetration/aspiration and nutritional modality: a cross-sectional study in patients with neurogenic dysphagia*. Acta Otorhinolaryngol Ital 2019 Mar 25. https://doi.org/10.14639/0392-100X-2136 [epub ahead of print].

Received: March 30, 2019 - Accepted: April 26, 2019

APPENDIX

With respect to our sample, the time spent to clear the 3 consistencies tested ranges from 4 up to 44 seconds. Stratifying linearly this time on 9 levels of 5 seconds, a factor of correction (FOC) is obtained to adjust the p-score in the following way: 0-5 secs =+ 1, 6-10 secs = +2, 11-15 secs = +3, 16-20 secs = +4, 21-25 secs = +5, 26-30 secs = +6, 31-35 secs = +7, 36-40 secs = +8, > 40 secs = +9 (Table V). The sum of the p-score total + FOC represents the timed p-score (tp-score). In this new role, the tp-score ranges from 5 up to 20, expressing itself as a continuum of severity [26]. The possibility of a clinical subdivision of the tp-score in further levels is under consideration.

Table V. Timed p-score (p-score).

Pooling	Endoscopic landmark	
Site	Vallecule – marginal zone	1
	Pyriform sinus	2
	Vestibule – vocal cords	3
	Below the vocal cords	4
Amount	Coating	1
	Minimum	2
	Maximum	3
Management	< 2	2
	2 > < 5	3
	> 5	4
Score	P 4-11	T
Time	FOC	X

Factor of correction (FOC): 0-5 secs =+1, 6-10 secs =+2, 11-15 secs =+3, 16-20 secs =+4, 21-25 secs =+5, 26-30 secs =+6, 31-35 secs =+7, 36-40 secs =+8, > 40 secs =+9

How to cite this article: Farneti D, Fattori B, Bastiani L. *Time as a factor during endoscopic assessment of swallowing: relevance in defining the score and severity of swallowing disorders.* Acta Otorhinolaryngol Ital 2019;39:244-249. https://doi.org/10.14639/0392-100X-N0221

Address for correspondence: Daniele Farneti, Audiology and Phoniatrics Service, Infermi Hospital, via Settembrini 2, 47923 Rimini, Italy. Tel/Fax. +39 0541 705146. E-mail: lele.doc@libero.it

ACTA OTORHINOLARYNGOLOGICA ITALICA 2019;39:250-256; doi: 10.14639/0392-100X-2173

OSAHS

Nasal pathologies in patients with obstructive sleep apnoea

Patologie nasali in pazienti affetti da sindrome delle apnee ostruttive del sonno

G. MAGLIULO[1], G. IANNELLA[1], A. CIOFALO[1], A. POLIMENI[2], M. DE VINCENTIIS[1], B. PASQUARIELLO[1], F. MONTEVECCHI[3], C. VICINI[3]

[1] Department of "Organi di Senso", University "Sapienza", Rome, Italy; [2] Department of Oral and Maxillo Facial Sciences, University "Sapienza", Rome, Italy; [3] Department of Head-Neck Surgery, Otolaryngology, Head-Neck and Oral Surgery Unit, Morgagni Pierantoni Hospital, Forlì, Italy

SUMMARY

Nasal obstruction is a frequent condition in patients with obstructive sleep apnoea (OSA). Nasal obstruction leads to mouth breathing, which is thought to destabilise the upper airway and aggravate the condition. Three conditions could be considered as the cause of the nasal breathing obstruction: anatomical conditions of the nose (septum deviation, hypertrophy of the inferior turbinates), chronic rhinosinusitis (CRS) and chronic nasal inflammation caused by allergic rhinitis or non-allergic cellular rhinitis. In this prospective study, we present an evaluation of all these possible rhino-sinusal aspects in OSA patients to correlate different nasal pathologies with nasal obstruction. Fifty patients with a diagnosis of OSA were enrolled in the study. In 70% of OSA patients, nasal obstruction was confirmed by clinical evaluation and rhinomanometry testing. Normal rhino-sinus aspects were present in only 20% of OSA patients, whereas one or more pathological rhino-sinus conditions were present in the remaining 80%. The percentage of OSA patients with a diagnosis of allergic rhinitis and non-allergic rhinitis was 18% and 26% respectively. Non-allergic rhinitis with neutrophils (NARNE) was the most frequent type of cellular rhinitis diagnosed in OSA patients (20% of cases). The results of the present study support and extend the observation that rhinitis is present in OSA patients. Mucosal inflammation caused by these conditions could be the cause of upper airway patency impairment inducing nasal mucosa swelling.

KEY WORDS: Obstructive sleep apnoea • Nasal obstruction • Allergic rhinitis • Non-allergic rhinitis • Nasal mucociliary clearance

RIASSUNTO

L'ostruzione nasale è una condizione frequentemente riportata in pazienti con sindrome delle apnee ostruttive del sonno (OSAS). L'ostruzione nasale porta alla respirazione orale, che si pensa possa destabilizzare le vie aeree superiori e aggravare l'OSAS. Tre condizioni potrebbero essere considerate come la causa dell'ostruzione respiratoria nasale: condizioni anatomiche nasali (deviazione del setto, ipertrofia dei turbinati inferiori), rinosinusite cronica (CRS) e infiammazione nasale cronica causata da rinite allergica o riniti non allergiche a cellularità. In questo studio prospettico presentiamo una valutazione di tutti questi possibili aspetti naso-sinusali in pazienti con OSAS al fine di correlare le diverse patologie nasali con l'ostruzione respiratoria nasale. Cinquanta pazienti con una diagnosi di OSAS sono stati arruolati nello studio. Nel 70% dei pazienti con OSAS, l'ostruzione nasale è stata confermata dalla valutazione clinica e dal test della respirazione con rinomanometria. Normali aspetti naso-sinusali erano presenti solo nel 20% dei pazienti con OSAS, mentre una o più condizioni patologiche naso-sinusali erano presenti nel restante 80%. L'incidenza di pazienti con OSAS con una diagnosi di rinite allergica e rinite non allergica era rispettivamente del 18% e del 26%. La rinite non allergica con neutrofili (NARNE) è stata la diagnosi più frequente nei pazienti con OSAS (20% dei casi). I risultati del presente studio supportano ed estendono l'osservazione che condizioni rinitiche sono presenti nei pazienti con OSAS. L'infiammazione della mucosa causata da queste condizioni potrebbe essere la causa della compromissione delle vie aeree superiori inducendo edema della mucosa nasale.

PAROLE CHIAVE: *Sindrome delle apnee ostruttive del sonno • Ostruzione nasale • Riniti allergiche • Riniti non allergiche • Clearance mucociliare nasale*

Introduction

Obstructive sleep apnoea (OSA) is a clinical entity characterised by recurring episodes of apnoea and/or hypopnoea during sleep due to a total or partial collapse of the upper airway [1-3]. OSA is characterised by night snoring with excessive daytime sleepiness and is commonly associated with a reduced quality of life, cardiovascular diseases, increased healthcare utilisation, motor vehicle accidents and decreased of cognitive performance [3-5]. Multilevel anatomical obstruction may play a role in OSA [3-7]. Nasal obstruction is frequently reported in these patients [5-8]. Various authors have supported the theory that nasal ob-

struction is a contributing factor in the pathogenesis of OSA despite numerous controversies [6-10]. Nasal obstruction leads to mouth breathing, which is thought to destabilise the upper airway and aggravate OSA [7-9]. Moreover, nasal breathing obstruction might represent a factor influencing the clinical history of this disease as well as the patient's compliance with CPAP [8-11].

What is the cause of nasal obstruction in OSA patients? Based on current knowledge about these patients, three conditions could be considered as the cause of the nasal breathing obstruction: anatomical conditions of the nose (septum deviation, hypertrophy of the inferior turbinates), chronic rhinosinusitis (CRS) and, as recently reported, nasal inflammation [5-13].

The effect of anatomical abnormalities of the nose on the sleep disorders has been analysed by different authors, who confirm an improvement in the number of apnoea/hypopnoea episodes following surgical correction of nasal abnormalities (septoplasty or turbinoplasty) [6-10]. It has also been well established that CRS causes impaired sleep quality and represents a risk factor for sleep apnoea and daytime sleepiness [14 15].

Recently, Zheng et al. [16] reviewed the literature and suggested a possible correlation between rhinitis and nasal obstruction in OSA patients. Today, it is generally accepted that the chronic nasal inflammation present in allergic and non-allergic rhinitis is a cause of increased nasal airway resistance due to mucosal swelling [9-13]. Therefore, nasal inflammation caused by these conditions may be a factor influencing nasal mucosa swelling and obstruction in OSA patients during sleep [10-13 16]. However, these conditions cannot be identified from clinical assessment, nasal symptom scores or rhinomanometry, and need to be studied using specific tests such as the skin prick test and nasal cytology [9-12 17].

In our opinion, the conditions responsible for nasal obstruction should not be considered separately in OSA patients but rather as a whole, since they may overlap and contribute in different ways to nocturnal nasal obstruction. No study has so far analysed the incidence of these different nasal conditions (AR, NAR, CRS) in OSA patients and their relationship to nasal obstruction. According to the above observations, we designed a prospective study with the subsequent aims: 1) perform an extensive evaluation of all rhino-sinusal aspects in OSA patients; 2) characterise the presence of allergic rhinitis (AR) and non-allergic cellular rhinitis. 3) correlate the different rhino-sinusal pathologies with nasal obstruction.

Materials and methods

Subjects

Patients affected by OSA were enrolled in our prospective clinical observational study at the "Organi di Senso" Department of the "Sapienza" University in Rome between December 2014 and January 2018.

Subjects eligible for the study were initially selected from patients referred to our Department with suspected OSA. All these patients underwent polysomnography (PSG) for diagnosis of this pathology. In accordance with the American Academy of Sleep Medicine (AASM), diagnosis and classification of OSA patients were performed on the basis of the apnoea + hypopnoea index (AHI) index. Patients were classified as normal (AHI was < 5/h), mild (AHI ≥ 5 and < 15 plus typical symptomatology), moderate (AHI ≥ 15 and < 30) and severe (AHI ≥ 30) [18 19].

Patients who had undergone surgery to the nose and/or rhino-sinusal surgery were excluded from the study. No patients were undergoing treatment with a CPAP device at home. Patients who were receiving topical or systemic steroids or other nasal therapies were also excluded from the study.

Clinical data, including height and weight, in order to calculate body mass index (BMI), medical history, tobacco use and a list of current medications were initially collected for each patient.

Nasal obstruction and possible presence of the three situations believed to cause OSA (nasal abnormalities, chronic sinusitis, allergic and non allergic cellular rhinitis) were investigated in all patients.

Nasal respiratory obstruction

The diagnosis of nasal respiratory obstruction was performed by clinical evaluation (self-reported sensation of nocturnal nasal obstruction and dry mouth upon awakening) and a rhinomanometry test. Anterior rhinomanometry is widely used to evaluate nasal resistance. It has been reported in the literature [20] that the mean total resistance in normal subjects ranges between 0.10 and 0.3 Pa/cm^3/s, with a mean of 0.25 Pa/cm^3/s. For this reason, total nasal airway resistance > 0.3 Pa/cm^3/s was considered pathological.

Nasal abnormalities

All patients were submitted to ENT physical examination with nasal endoscopy (2.7 mm 0° rigid endoscope) to evaluate the features of nasal structures (septum deviation, hypertrophy of the inferior turbinates) and detect

anatomical abnormalities responsible for nasal obstruction.

Chronic rhinosinusitis

A diagnosis of CRS was made according to the EPOS classification of rhinosinusitis [21] which considers patients with two or more signs and symptoms (bilateral nasal obstruction, nasal discharge, facial pain/ headache, subjective olfactory dysfunction) for 12 or more weeks, without complete resolution. Any rhinosinusal pathologies (such as nasal polyposis, aspects of chronic rhinosinusitis, nasal infection) were also investigated during nasal endoscopy.

Moreover, all patients underwent CT scan of rhinosinusal structures (axial, coronal and sagittal projections, without the use of intravenous contrast) at the same time as the ENT examination to confirm or exclude CRS.

Allergic and non-allergic cellular rhinitis (AR and NAR)

According to ARIA diagnostic criteria [22] patients with at least 2 of the allergic rhinitis (AR) symptoms were investigated for AR (suggestive symptomatology according to Skin prick test/IgE positivity) and non-allergic cellular rhinitis (NAR). Cellular rhinitis was diagnosed by nasal cytology in patients with a clinical suggestive history of AR, but without allergic sensitisation (skin prick test/IgE positivity) [11 22-24].

Patients with rhinopathy were subdivided on the basis of the prick test and of nasal cytology into subjects with AR or with NAR [11 22-24]. Cellular forms were further sub-divided on the basis of cytotype [11 24].

In accordance with the European Academy of Allergy and Clinical Immunology, standardised allergen panels were employed to detect an IgE-mediated allergic response in all CVID patients [22 23]. The allergen panel consisted of the following: house dust mites (Dermatophagoides farinae and pteronyssinus), cat and dog hair, grasses mix, composite mix, parietaria judaica, birch, hazel tree, olive tree, alternaria tenuis, cladosporium and aspergilli mix; the concentration of allergen extracts was 100 IR/mL (Stallergenes, Milan, Italy). Sensitisation was considered present when the diameter of local reaction was equal to or greater than 3 mm [22 23].

Scrapings of the nasal mucosa were placed on a microscope slide, fixed for air dry and stained by the May-Grunwald-Giemsa method [11 24]. The semi-quantitative analysis proposed by Meltzer et al. [25] was used to evaluate nasal cytology.

According to the semi-quantitative analysis and to the results of skin prick tests, patients were classified as normal or as having AR or NAR [11 22-25]. NAR subjects, also defined as cellular rhinitis, were further subdivided into NARNE (neutrophils > 50% with absent spores and bacteria); NARES (eosinophils > 20%); NARMA (mast cells > 10%); NARESMA (eosinophils > 20% and mast cells > 10%) [11 24 25].

Diagnosis of rhino-sinusal pathologies

By comparing the results of the above mentioned tests, different conditions were diagnosed: isolated nasal abnormalities, chronic rhinosinusitis (CRS),AR), AR + CRS, NAR and NAR + CRS. Each of these conditions was correlated with the presence of nasal obstruction.

Statistical analysis

A Student's T test and χ^2 test were employed to evaluate the significance of multiple factors. A p value of < 0.05 was taken as the threshold of statistical significance. This research study was performed in accordance with the principles of the Declaration of Helsinki and approved by the local Ethics Committee of the University ''Sapienza'' of Rome. All patients gave written informed consent for the PSG, CT scan and other rhinologic tests of the study.

Results

55 patients with a diagnosis of OSA were enrolled in the study. The mean age of the study group was 55.2 years (range 35-79): 33 subjects were male and 17 were female. The clinical characteristics of the study group are reported in Table I.

In 35 (70%) patients with OSA, nasal obstruction was confirmed by clinical evaluation and rhinomanometry testing (Table I). No difference emerged between OSA subgroups and the incidence of nasal obstruction using the chi square test ($p > 0.5$ in each case). This was confirmed by regression analysis between AHI index and mean nasal airway resistance calculated with the rhinomanometry test ($p = 0.7$) (Fig. 1).

The diagnostic protocol used in this study showed that normal rhino-sinus aspects were present in only 20% of OSA patients, whereas one or more pathological rhino-sinus conditions were present in the remaining 80%.

The incidence of the different rhino-sinus pathologies present in OSA patients is reported in Table II. Interestingly, the incidence of OSA patients with a diagnosis of AR and NAR was 18% and 26%, respectively.

In patients with a diagnosis of AR or NAR, mean AHI did not differ from those without rhinitis diagnosis ($p > 0.05$ in each case; Fig. 2).

In 82.5% of patients suffering from a pathological rhino-sinus condition, nasal obstruction was present. There was

Table I. Clinical characteristics of the study group.

	OSA patients 50 pts	
Mean age	55.2 years (range 35-79 years old) Mild OSA: 54.5 Moderate OSA: 55.1 Severe OSA: 57.4	
Sex	33 male 17 female	
BMI (mean value)	32.5	
OSA severity	Mild OSA 15 pts (mean AHI = 10.9) Moderate OSA 18 pts (mean AHI = 21.5) Severe OSA 17 pts (mean AHI = 42.6)	
NASAL OBSTRUCTION (Clinical evaluation + rinomanometry)	35 pts	70%
Mild OSA	10 pts	20%
Moderate OSA	11 pts	22%
Severe OSA	14 pts	28%

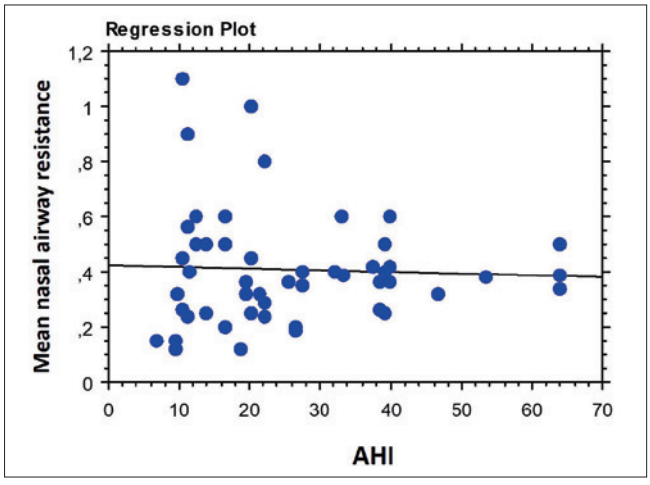

Fig. 1. Regression analysis between AHI index and mean nasal airway resistance calculated with the rhinomanometry test (p = 0.7).

a significant difference between normal and pathological subjects regarding nasal obstruction (p = 0.004).

Analysing the different pathological rhino-sinus conditions regarding the simultaneous presence or absence of nasal obstruction, interesting aspects emerged (Table II): 100% of patients with AR, CRS and AR+CRS showed nasal obstruction, whereas this condition was present in 92.3% of patients suffering from NAR. Different from these inflammatory conditions, only 60% of patients with isolated nasal abnormalities (nasal deviation, inferior turbinate hypertrophy etc.) had a diagnosis of nasal obstruction.

Table III displays a subdivision of different pathological rhino-sinus conditions according to Nasal Cytological Outcomes classification. Non-allergic rhinitis with neu-

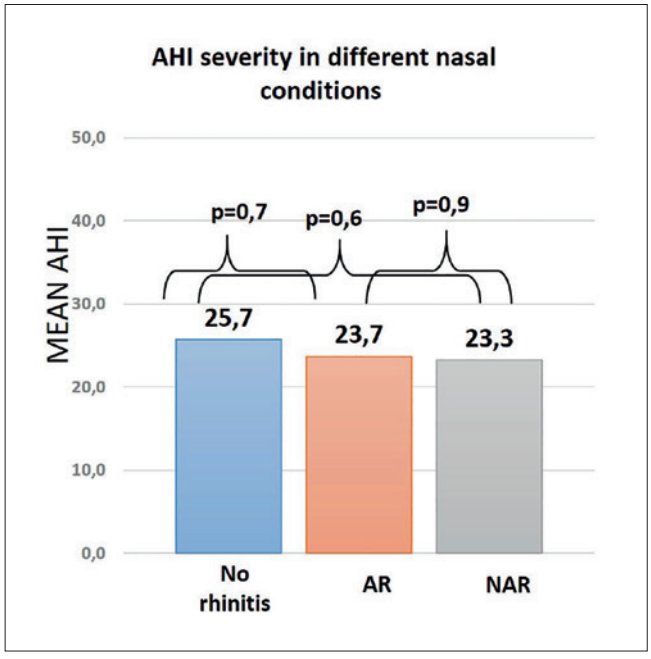

Fig. 2. Mean AHI value of patients without rhinitis and patients with allergic rhinitis and non-allergic rhinitis diagnosis.

Table II. Different rhino-sinusal pathologies present in OSA patients.

	OSA patients N (%)	OSA patients with nasal obstruction N (%)	OSA patients without nasal obstruction N (%)
Number of patients	50	35 (70%)	15 (30%)
Normal rhino-sinusal aspects	10 (20%)	2 (20%)	8 (80%)
Pathological rhino-sinusal aspects	40 (80%)	33 (82.5%)	7 (15.5%)
Isolated nasal abnormalities (nasal septum deviation, inferior turbinate hypertrophy)	15 (30%)	9 (60%)	6 (40%)
Chronic rhinosinusitis (CRS)	3 (6%)	3 (100%)	-
Allergic rhinitis (AR)	4 (8%)	4 (100%)	-
CRS + AR	5 (10%)	5 (100%)	-
Non-allergic rhinitis (NAR)	13 (26%)	12 (92.3%)	1 (7.7%)
NAR + CRS	-	-	-

Table III. Nasal cytological outcomes.

Nasal cytology	Cytological characteristics	Total of patients N (%)
Allergic rhinitis (skin prick test +)	Numerous neutrophils and eosinophils, partially degranulated	9 (18%)
Non allergic cellular rhinitis		13 (26%)
NARNE	Neutrophils > 50% with absent spores and bacteria	10 (20%)
NARMA	Mast cells > 10%	-
NARES	Eosinophils > 20%	2 (4%)
NARESMA	Eosinophils > 20% and mast cells > 10%	1 (2%)
Infective*	Numerous neutrophils and bacteria	3 (6%)

**: patients with cytological characteristics of infective rhinitis was the same with a CRS diagnosis.*

trophils (NARNE) was the most frequent NAR diagnosed in OSA patients (20% of cases). In patients diagnosed with AR, numerous neutrophils and eosinophils, partially degranulated, were evident at cytology, whereas in all those diagnosed with CRS cytological characteristics of infective rhinitis were present.

Discussion

There is evidence showing that nasal obstruction can contribute to the pathogenesis of OSA, decrease the quality of life in OSA patients, contribute to snoring and represent an obstacle for effective treatment with CPAP [6-11 26-28].

What is the cause of nasal obstruction in OSA patients? A systematic evaluation of nasal obstruction remains challenging due to the high number of factors that contribute to nasal obstruction [6 8 10]. Currently, nasal obstruction evaluation in OSA patients in most medical environments is limited to anterior rhinoscopy: this allows evaluation of anterior septal deviation, internal nasal valve angle and inferior turbinate size, but fails to identify other factors that could play a role in nasal obstruction [6-11]. In addition, characteristics of the anterior nasal cavity frequently do not correlate with the patient's symptoms and/or nasal breathing. Patients often complain of nasal obstruction despite no objective signs of anatomical abnormalities in the nasal cavity when examined with anterior rhinoscopy and/or nasal endoscopy [11 16].

Other aetiologies for nasal obstruction such as chronic sinusitis or inflammatory problems such as AR and/or non-allergic cellular rhinitis, could be the reason why nocturnal nasal breathing is absent [11 16].

In our observational study, it emerged that 70% of OSA patients displayed nasal respiratory obstruction. Normal rhino-sinusal aspects were present in only 20% of OSA patients examined, whereas one or more pathological rhino-sinusal conditions were present in 80% of enrolled patients. It is interesting to note that only 30% of these patients had isolated anatomical abnormalities, 6% had chronic rhinosinusitis, 18% AR and 26% NAR. These corroborate the results reported by Shadan et al. [29] who employed nasal cytology as a marker of clinically silent inflammation in a group of 38 OSA patients, diagnosing AR in 37% and NAR in 21% of subjects.

Regarding the cause of nasal obstruction in OSA patients, it should be noted that 95.4% of patients with nasal inflammatory conditions such as AR or NAR, suffered from nocturnal respiratory obstruction. In contrast, 40% of patients with isolated nasal abnormalities did not suffer from nasal obstruction.

Some observational studies have shown how the nasal congestion induced by AR is an important factor in sleep impairment [12 16 30]. One large population-based study involving about 5,000 adults demonstrated that individuals reporting frequent nasal congestion (5 nights/month) due to allergy were almost twice as likely to have moderate-to-severe SDB than individuals without nasal congestion due to allergy [15]. Moreover, the incidence of AR among patients with OSA was recently estimated to be between 11.7% and 27.1% with no significant differences in sleeping parameters between allergic and non-allergic patients [10].

The mechanism through which AR causes poor quality of sleep and daytime fatigue is not entirely clear, but several factors are believed to be involved. Inflammatory mediators such as interferon- (IFN-) gamma, tumour necrosis factor- (TNF-) alpha, interleukin- (IL-) 1b, IL-4, IL-10, postural changes and certain therapeutic agents, such as antihistamines, may have a direct impact on sleep regulation [12 13 16]. However, like AR, non-allergic cellular rhinitis should also be considered among the possible causes of nasal congestion due to inflammation of the mucosa [11-13].

A high incidence of NAR of 28.7% in OSA has been recently reported by Zheng et al. [12] using validated questionnaires and skin prick tests. The patients in this study with NAR had lower average arterial oxygen saturation and minimal arterial oxygen saturation, compared with subjects categorised as no-rhinitis.

According to the recent ARIA classification, in order to obtain a more precise classification of NAR, nasal cytology tests should be performed in patients with suggestive history of AR, but without allergic sensitisation (Skin prick test/IgE positivity) [11 22-24].

Using these tests, Gelardi et al. [11] were the first to demonstrate a subclinical nasal inflammation in patients with OSA in CPAP treatment. They showed that at cytology examination 9 patients (28.1%) had aspects of NARNE, 6 (18.7%) of NARES and 4 (12.5%) of NARESMA, whereas 5 (15.6%) patients showed cellular signs of AR with numerous neutrophils and eosinophils, partially degranulated: finally in 8 (25%) patients the cytologic signs of rhinosinusitis characterised by numerous neutrophils and the presence of bacteria. Using the same diagnostic protocol as Gelardi et al. [11], in our study group of OSA patients NARNE was present in 20% of cases, whereas NARES and NARESMA was seen in 4% and 2%, respectively.

Different from other studies, patients with a diagnosis of AR or NAR did not seem to have a mean AHI that was significantly different from that of patients not diagnosed with rhinitis [12 16] ($p > 0.05$ in all cases). Similar results have been reported by Kramer et al. [10] who found no significant differences in sleeping behaviour or polysomnography parameters comparing allergic and non-allergic patients.

The results of the present study support and extend the observation that rhinitis is present in OSA patients [11 12 16]. Mucosa inflammation caused by these conditions may further compromise upper airway patency by inducing nasal mucosa swelling.

In our opinion, inflammatory conditions such as AR and/or cellular rhinitis should be evaluated in OSA patients to avoid unnecessary surgical procedures, re-establish normal nasal function and improve compliance with eventual CPAP treatment [11 29-32]. Notwithstanding, further studies are necessary to investigate inflammation mediators in nasal mucosa of OSA patients and correlate these with the presence of nasal obstruction and the severity of OSA.

Conclusions

One or more pathological rhino-sinusal conditions were present in 80% of OSA patients evaluated in this study. The incidence of OSA patients with a diagnosis of AR and NAR was 18% and 26%, respectively. A diagnosis of AR or NAR does not seem to correlate with severity of AHI.

Conflict of interest statement

None declared.

References

1. Bosi M, De Vito A, Kotecha B, et al. *Phenotyping the pathophysiology of obstructive sleep apnea using polygraphy/polysomnography: a review of the literature.* Sleep Breath 2018;22:579-92. https://doi.org/10.1007/s11325-017-1613-3.
2. Paskhover B. *An introduction to obstructive sleep apnea.* Otolaryngol Clin North Am 2016;49:1303-6. https://doi.org/10.1016/j.otc.2016.07.007.
3. Kapur VK. *Obstructive sleep apnea: diagnosis, epidemiology, and economics.* Respir Care 2010;55:1155-67.
4. Pinto JA, Ribeiro DK, Cavallini AF, et al. *Comorbidities associated with obstructive sleep apnea: a retrospective study.* Int Arch Otorhinolaryngol 2016;20:145-50. https://doi.org/10.1055/s-0036-1579546.
5. Schwartz RN, Payne RJ, Forest VI, et al. *The relationship between upper airway collapse and the severity of obstructive sleep apnea syndrome: a chart review.* J Otolaryngol Head Neck Surg 2015;44:32. https://doi.org/10.1186/s40463-015-0086-2.
6. Migueis DP, Thuler LC, Lemes LN, et al. *Systematic review: the influence of nasal obstruction on sleep apnea.* Braz J Otorhinolaryngol 2016;82:223-31. https://doi.org/10.1016/j.bjorl.2015.05.018.
7. Alwani A, Rubinstein I. *The nose and obstructive sleep apnea.* Curr Opin Pulm Med 1998;4:361-2.
8. Vicini C, De Vito A, Benazzo M, et al. *The nose oropharynx hypopharynx and larynx (NOHL) classification: a new system of diagnostic standardized examination for OSAHS patients.* Eur Arch Otorhinolaryngol 2012;269:1297-300. https://doi.org/10.1007/s00405-012-1965-z.
9. Georgalas C. *The role of the nose in snoring and obstructive sleep apnoea: an update.* Eur Arch Otorhinolaryngol 2011;268:1365-73. https://doi.org/10.1007/s00405-010-1469-7.
10. Kramer MF, De La Chaux R, Dreher A, et al. *Allergic rhinitis does not constitute a risk factor for obstructive sleep apnea syndrome.* Acta Otolaryngol 2001;121:494-9.
11. Gelardi M, Carbonara G, Maffezzoni E, et al. *Regular CPAP utilization reduces nasal inflammation assessed by nasal cytology in obstructive sleep apnea syndrome.* Sleep Med 2012;13:859-63. https://doi.org/10.1016/j.sleep.2012.04.004.
12. Zheng M, Wang X, Ge S, et al. *Allergic and non-allergic rhinitis are common in obstructive sleep apnea but not associated with disease severity.* J Clin Sleep Med 2017;13:959-66. https://doi.org/10.5664/jcsm.6694.
13. Passali D, Corallo G, Petti A, et al. *A comparative study on oxidative stress role in nasal breathing impairment and obstructive sleep apnoea syndrome.* Acta Otorhinolaryngol Ital 2016;36:490-5. https://doi.org/10.14639/0392-100X-1361.
14. Jiang RS, Liang KL, Hsin CH, et al. *The impact of chronic rhinosinusitis on sleep-disordered breathing.* Rhinology 2016;54:75-9. https://doi.org/10.4193/Rhin15.204.
15. Young T, Finn L, Kim H. *Nasal obstruction as a risk factor for sleep-disordered breathing. The University of Wisconsin Sleep and Respiratory Research Group.* J Allergy Clin Immunol 1997;99:S757-62.
16. Zheng M, Wang X, Zhang L. *Association between allergic and nonallergic rhinitis and obstructive sleep apnea.* Curr Opin Allergy Clin Immunol 2018;18:16-25. https://doi.org/10.1097/ACI.0000000000000414.
17. Passali FM, Bellussi L, Mazzone S, et al. *Predictive role of nasal functionality tests in the evaluation of patients before nocturnal polysomnographic recording.* Acta Otorhinolaryngol Ital 2011;31:103-8.
18. Berry RB, Brooks R, Charlene E, et al. *AASM Manual for the scor-*

ing of sleep and associated events. American Academy of Sleep Medicine; 2015.

19 Epstein LJ, Kristo D, Strollo PJ Jr, et al. *Clinical guideline for the evaluation, management and long-term care of obstructive sleep apnea in adults.* J Clin Sleep Med 2009;5:263-76.

20 Merkle J, Kohlhas L, Zadoyan G, et al. *Rhinomanometric reference intervals for normal total nasal airflow resistance.* Rhinology 2014;52:292-9. https://doi.org/10.4193/Rhin13.220.

21 Fokkens WJ, Lund VJ, Mullol J, et al. *EPOS 2012: European position paper on rhinosinusitis and nasal polyps 2012. A summary for otorhinolaryngologists.* Rhinology 2012;50:1-12. https://doi.org/10.4193/Rhino50E.

22 Brożek JL, Bousquet J, Agache I. *Allergic rhinitis and its impact on asthma (ARIA) guidelines-2016 revision.* J Allergy Clin Immunol 2017;140:950-8. https://doi.org/10.1016/j.jaci.2017.03.050.

23 Heinzerling L, Mari A, Bergmann K, et al. *The skin prick test - European standard.* Clin Transl Allergy 2013;3:3. https://doi.org/10.1186/2045-7022-3-3.

24 Gelardi M. *Atlas of nasal cytology.* Milano: Edi Ermes; 2012.

25 Meltzer EO, Orgel HA, Jalowayski AA. *Histamine levels and nasal cytology in children with chronic otitis media and rhinitis.* Ann Allergy Asthma Immunol 1995;74:406-10.

26 Magliulo G, de Vincentiis M, Iannella G, et al. *Olfactory evaluation in obstructive sleep apnoea patients.* Acta Otorhinolaryngol Ital 2018;38:338-45. https://doi.org/10.14639/0392-100X-1981.

27 Bican A, Kahraman A, Bora I, et al. *What is the efficacy of nasal surgery in patients with obstructive sleep apnea syndrome?* J Craniofac Surg 2010;21:1801-6. https://doi.org/10.1097/SCS.0b013e3181f40551.

28 Magliulo G, de Vincentiis M, Iannella G, et al. *Eustachian tube evaluation in patients with obstructive sleep apnea syndrome.* Acta Otolaryngol 2018;138:159-64. https://doi.org/10.1080/00016489.2017.1385846.

29 Shadan FF, Jalowayski AA, Fahrenholz J. *Nasal cytology: a marker of clinically silent inflammation in patients with obstructive sleep apnea and a predictor of noncompliance with nasal CPAP therapy.* J Clin Sleep Med 2005;1:266-70.

30 Thompson A, Sardana N, Craig TJ. *Sleep impairment and daytime sleepiness in patients with allergic rhinitis: the role of congestion and inflammation.* Ann Allergy Asthma Immunol 2013;111:446-51. https://doi.org/10.1016/j.anai.2013.05.020.

31 Fiorita A, Scarano E, Mastrapasqua R, et al. *Moderate OSAS and turbinate decongestion: surgical efficacy in improving the quality of life and compliance of CPAP using Epworth score and SNOT-20 score.* Acta Otorhinolaryngol Ital 2018;38:214-21. https://doi.org/10.14639/0392-100X-1935.

32 Toraldo DM, Passali D, Sanna A, et al. *Cost-effectiveness strategies in OSAS management: a short review.* Acta Otorhinolaryngol Ital 2017;37:447-53. https://doi.org/10.14639/0392-100X-1520.

Received: April 9, 2018 - Accepted: May 24, 2018

How to cite this article: Magliulo G, Iannella G, Ciofalo A, et al. *Nasal pathologies in patients with obstructive sleep apnoea.* Acta Otorhinolaryngol Ital 2019;39:250-256. https://doi.org/10.14639/0392-100X-2173

Address for correspondence: Giuseppe Magliulo, Department of "Organi di Senso", University "Sapienza", via Gregorio VII 80, 00165 Rome, Italy. E-mail: giuseppemagliuloorl@yahoo.com

Audiology

Hearing loss in very preterm infants: should we wait or treat?

L'ipoacusia nei neonati estremamente prematuri: trattare subito o aspettare?

S. FREZZA[1], P. CATENAZZI[1], R. GALLUS[2], F. GALLINI[1], M. FIORETTI[1], R. ANZIVINO[2], M. CORSELLO[1], F. COTA[1], G. VENTO[1], G. CONTI[2]

[1] Department of Woman and Child Health, Division of Neonatology, [2] Department Head and Neck Surgery, Institute of Otorhinolaryngology, Università Cattolica del Sacro Cuore, Fondazione Policlinico Universitario A. Gemelli IRCSS, Rome, Italy

SUMMARY

This study investigated hearing threshold changes during the first year of corrected age (CA) in infants admitted in a neonatal intensive care unit (NICU). In 5 years, 239 infants with birth weight (BW) ≤ 1,000 gm and/or gestational age (GA) ≤ 30 weeks were enrolled. Hearing was evaluated by oto-acoustic emission (OAEs) before discharge and auditory brainstem response (ABR) within 3 months of CA. Infants affected by unilateral or bilateral hearing loss were addressed to audiological follow-up until definitive diagnosis (within 6 months of CA). Changes in hearing threshold were also carefully analysed. 207 (86.6%) infants had normal hearing while 32 infants (13.4%) showed hearing loss (HL) at the confirmative ABR evaluation (9 mild, 16 moderate, 4 severe, 3 profound). The latter showed lower GA (27.7 ± 2 vs 28.4 ± 1.2; p = 0.0061) and BW (950 ± 390 vs 1,119 ± 326 gm; p = 0.0085). At final evaluation, 15 infants (47%) recovered a normal hearing. HL was confirmed in 17 patients. Among these, 3 infants were addressed to audiological follow-up (one case of mild unilateral hearing loss (UHL) and two with moderate UHL), while in 14 cases (44%) with bilateral sensory neural hearing loss (SNHL) (7 moderate, 4 severe, 3 profound) hearing aids were prescribed. They showed significantly lower GA and longer hospital stay in the NICU in comparison with infants without indication for audiological habilitation (18 infants) (GA 26.2 ± 2.2 weeks vs 28.4 ± 2.4; p = 0.01; NICU stay 132 ± 67 vs 59 ± 7; p = 0.0002). Definitive diagnosis was obtained at 5.9 ± 1.3 months of CA. Our study confirms the importance of audiological surveillance in preterm newborns. Hearing thresholds of preterm infants with hearing loss can change during the first year of CA and we observed normalisation in 47% of our patients. Most vulnerable to permanent SNHL were very preterm infants with a longer NICU stay, while a shorter stay represents a favourable prognostic factor for hearing improvement.

KEY WORDS: Preterm infants • NICU • Sensorineural hearing loss • Newborn Hearing Screening • Hearing Aids

RIASSUNTO

Un'ipoacusia permanente infantile (IPI) può avere gravi conseguenze sullo sviluppo del linguaggio e delle abilità cognitive. Le IPI congenite hanno una prevalenza di circa 1,5-3 nuovi casi per mille neonati. Esistono tuttavia alcuni gruppi di bambini, come quelli ricoverati in unità di terapia intensiva neonatale (UTIN), in cui il rischio può essere 10-20 volte maggiore. La diagnosi precoce delle IPI consente di adottare misure altamente efficaci di trattamento/abilitazione. Non vi è, tuttavia, uniformità nella gestione del follow-up di questi bambini, con difficoltà nella definizione fisiopatologica del deficit uditivo e nella sua quantificazione. Scopo dello studio è stato quello di valutare la prevalenza delle ipoacusie e i risultati del follow-up audiologico in una popolazione di prematuri dimessi dalla UTIN. Sono stati inclusi nello studio i neonati con EG ≤ 30 settimane e PN ≤ 1.000 gr, nati nell'arco di 5 anni, dimessi dalla UTIN e seguiti presso il nostro Servizio di Follow-Up. Tutti sono stati sottoposti a registrazione di OAEs alla dimissione (dopo le 32 settimane di età post-mestruale) e ABR diagnostico (con strumenti e personale di laboratorio) entro i tre mesi di età corretta (EC). Tutti i bambini con ipoacusia mono- o bilaterale da lieve (soglia elettrofisiologia oltre 20, entro 40 dBnHL) a profonda (soglia > 90 dBnHL) sono stati controllati fino alla diagnosi audiologica definitiva, (entro i 5-6 mesi di EC) e sono state verificate, in particolare, le eventuali modificazioni della soglia elettrofisiologica entro i 12 mesi di EC. Di 239 bambini valutati, 32 (13,4%; EG 27 ± 2 sett, PN 950 ± 390 g) hanno presentato un'ipoacusia mono/bilaterale (9 lievi, 16 medie, 4 gravi, 3 profonde) ai tre mesi di EC. I 32 bambini con rilievo iniziale di ipoacusia hanno mostrato nei controlli successivi una normalizzazione della soglia uditiva in 15 casi (47%) e in 3 casi un'ipoacusia monolaterale (1 caso lieve, 2 casi media) che non ha richiesto protesizzazione. In 14 casi (44%) (EG 26,2 ± 2,2 sett, PN 820 ± 330 g) è stata confermata una diagnosi di ipoacusia neurosensoriale bilaterale (7 medie, 4 gravi, 3 profonde). In questi ultimi la diagnosi di conferma è stata completata entro il 5°mese di EC, con protesizzazione acustica, attuata in media a 5,9 (± 1,3 mesi) di EC. Lo studio conferma l'importanza dell'intervento audiologico nella gestione dei neonati estremamente prematuri. Le soglie uditive dei neonati pretermine con ipoacusia possono cambiare durante il primo anno di EC e noi abbiamo descritto una normalizzazione nel 47% dei nostri pazienti. Maggiormente predisposti all'IPI erano i neonati estremamente pretermine con una permanenza in UTIN più lunga, mentre una durata inferiore del ricovero si è rivelata essere un fattore prognostico favorevole per il miglioramento della soglia uditiva.

PAROLE CHIAVE: *Neonati pretermine • UTIN • Ipoacusia neurosensoriale • Screening uditivo neonatale • Protesi acustiche*

Introduction

Permanent bilateral hearing loss (PHL) affects 1-3/1000 live births in wellborn infants and 2-4/100 infants in the neonatal intensive care unit (NICU) population [1-3]. In order to achieve effective treatment, congenital or perinatal hearing loss should be recognised within three months of life, with confirmative audiological diagnosis and early intervention before the 6th month of age [4]. Early treatment is essential, as the first year of life is critical for normal development of speech and language, as well as intellectual and emotional growth [5-7].

Preterm infants, with an increased risk for SNHL and auditory neuropathy spectrum disorders, are screened with auditory brainstem response (ABR) which allows objective and accurate assessment of the hearing function, with normal variation according to age due to physiological maturation of the auditory pathway [7].

In the last few years, an improvement over time with regards to initial hearing thresholds in infants who failed newborn hearing screening has been reported in several studies [1 2 8 9]. Changes in hearing threshold in infants can depend on factors temporary affecting the auditory periphery as well the neural pathway and/or on a delay in "auditory maturation" [9-11]. This latter factor seems to have a major role in premature infants, whose behaviour has been addressed in studies based on both heterogeneous [1 2] or homogeneous [12-14] groups of children.

Most of these studies lack uniformity with regards to the degree of prematurity, age of the first audiological diagnosis, definition of the hearing loss level and procedures and methodology of follow-up. In addition, they are usually based on retrospective/descriptive analysis of small series of infants.

This study reports the prevalence rates of SNHL in a cohort of preterm infants admitted in the NICU of "A. Gemelli" Hospital in Rome. We prospectively analysed changes in threshold during the first 12 months of CA in infants who resulted affected by hearing loss at initial evaluation on the basis of homogeneously applied objective procedures.

Materials and methods

From January 2009 to December 2014, 245 infants with birth weight (BW) ≤ 1,000 gm and/or GA ≤ 30 weeks who were treated in our NICU were included in the study. These infants were enrolled prospectively in our follow-up monitoring, within the first weeks after discharge. Multidisciplinary health assessments occurred at 40 weeks of post menstrual age (PMA) and at 3, 6, 9 and 12 months of CA.

A schematic overview of the neonatal hearing screening program in our Division for NICU infants is shown in Figure 1. For the purpose of the present study, all infants underwent a diagnostic ABR recording at about 3 months (+ 2 weeks) of CA. Infants affected by hearing impairment where then addressed, within 5-6 months of CA, to confirmative audiological evaluation, which was aimed to eventually activate the rehabilitation programme. Hearing impaired children were further evaluated about every three months within the first year of CA.

The audiological evaluation consisted of history, otoscopy, oto-acoustic emission (OAEs), diagnostic ABR and tympanometry.

Infants affected by bilateral hearing loss were also evaluated with behavioural audiometry (behavioural observation audiometry or visual reinforcement audiometry, depending on the child's age and participation) after confirmative physiological diagnosis and fitting of hearing aids. Results of behavioural evaluation are not considered in the present study.

ABRs were recorded in a soundproof and electrically shielded room. All children were in natural sleep throughout the recording session. Both ears were sequentially tested. Stimuli were clicks 0.1 msec duration and alternating polarity, presented by earphones (TDH-49P) at 21.1/sec. ABRs were recorded using the ICS Chartr EP system, with an ICS Chartr PA-800 preamplifier. Surface

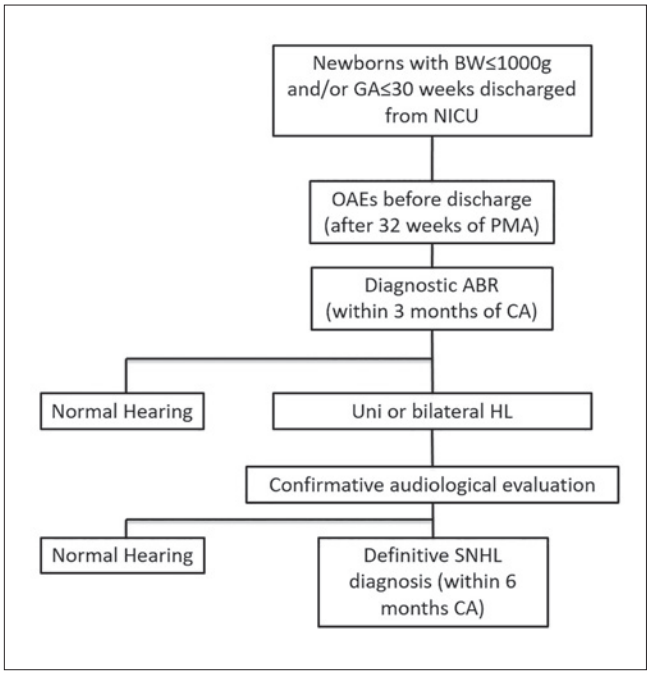

Fig. 1. Schematic overview of the neonatal hearing screening programme for preterm infants in "A. Gemelli" Hospital.

electrodes where placed at the ipsilateral ear lobe (-), vertex (+), or just ventrally to the bregmatic fontanelle, and contralateral ear lobe (ground) and impedance was kept under 5 kΩ. The signal was amplified (100 k) and filtered (50-3,000 Hz). Each trace was obtained by averaging 500-1,500 single epochs and was replicated at least twice, mainly at the electrophysiological threshold level. This was determined as the lowest intensity level where a response could be assessed thorough the identification of the V wave, starting from 60 dB nHL, by a "20 dB down - 10-5 dB up" procedure. ABR measurements were performed by an audiology technician under supervision of an expert physician. Our protocol did not include automatic ABR.

TEOAEs measurements were performed using the Madsen Accuscreen®, which performs an evaluation of a patient's TEOAES through a noise-weighted averaging and counting of significant signal peaks. The stimulus sent through the probe is a non-linear click sequence at 60 Hz rate with a sound pressure level of 70-84 dB SPL with a self calibration depending on ear canal volume.

Tympanometry was performed with a Tympstar® by Granson Stadler with a 226/660 (depending on the age) Hz probe tone (85 dB SPL ± 1.5 dB).

Changes in hearing threshold were particularly analysed. Hearing loss was defined as: mild (20-40 dB nHL), moderate (41-70 dB nHL), severe (71-90 dB nHL), profound (> 90 dB nHL) [4].

Parents were supplied with detailed informing brochures about significance and importance of the path of audiological screening and rehabilitation, when needed.

With regard to terminology used to define the age of patients, the standardised definition of gestational age (GA), postmenstrual age (PMA) and corrected age (CA) was applied as reported by Engle in 2004 [15].

For statistical analysis, continuous variables were presented as mean ± standard deviation (SD) and categorical variables as number and percentage. Comparisons between continuous variables were performed using Student's t-test. Comparisons between categorical variables were performed using the Fisher's exact test. A two-tailed $p < 0.05$ was considered significant. Statistical analyses were performed with Graph Pad Prism 4 software.

Results

Among the 245 infants enrolled, only 6 (2.4%) were lost to follow up.

At the initial ABR 207 of 239 (86.6%) subjects showed normal hearing thresholds, while 32 (13.4%) showed unilateral or bilateral HL (9 mild, 16 moderate, 4 severe, 3 profound HL). Infants with hearing impairment showed a significantly lower GA (27 ± 2 vs 28.4 ± 1.2 weeks; $p = 0.0061$) and BW (950 ± 390 vs 1,119 ± 326 g; $p = 0.0085$) in comparison to normal hearing babies.

Substantial changes in hearing threshold were observed during follow-up. Fifteen of 32 children with an initial finding of bilateral HL (47%) showed a normalisation of hearing threshold evaluations (Fig. 2). Figure 3 displays the changes in ABR in one of these infants. From an initial bilateral moderate/severe SNHL, there was a clear progressive improvement of hearing threshold. In particular, at the first ABR (performed at 40 weeks of PMA) the electrophysiological threshold was 60/50 dB nHL (right side) and 80 dB nHL (left side). When the baby was 3 months of CA, the threshold was unchanged on the right but showed a remarkable improvement on the left (30 dB nHL). In the following evaluation at 6 months of CA a substantial improvement of hearing was observed bilaterally, with threshold near to normalisation (left ear 30 dB nHL, right ear 20 dB nHL).

To better appreciate the effective improvement pattern of these 15 infants, their 30 ears were evaluated separately. Hearing threshold improved from a moderate loss to a normal level in 12 ears (40%) and from a mild loss to normal hearing in 18 ears (60%). The average threshold variation was 20 ± 10.94 dB nHL (range 10-45 dB nHL). Normalisation of hearing threshold was reached at the

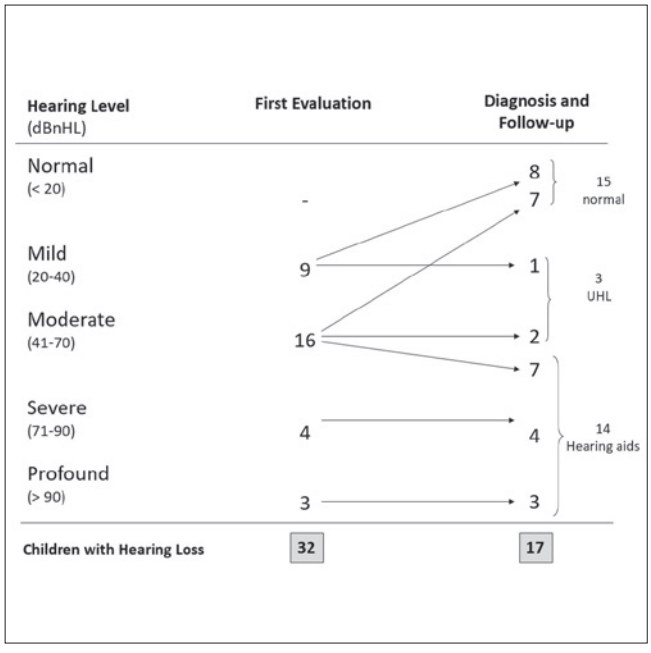

Fig. 2. Changes in hearing thresholds in our population at the time of first evaluation (ABR within 3 months of CA) and during hearing diagnosis and follow-up.

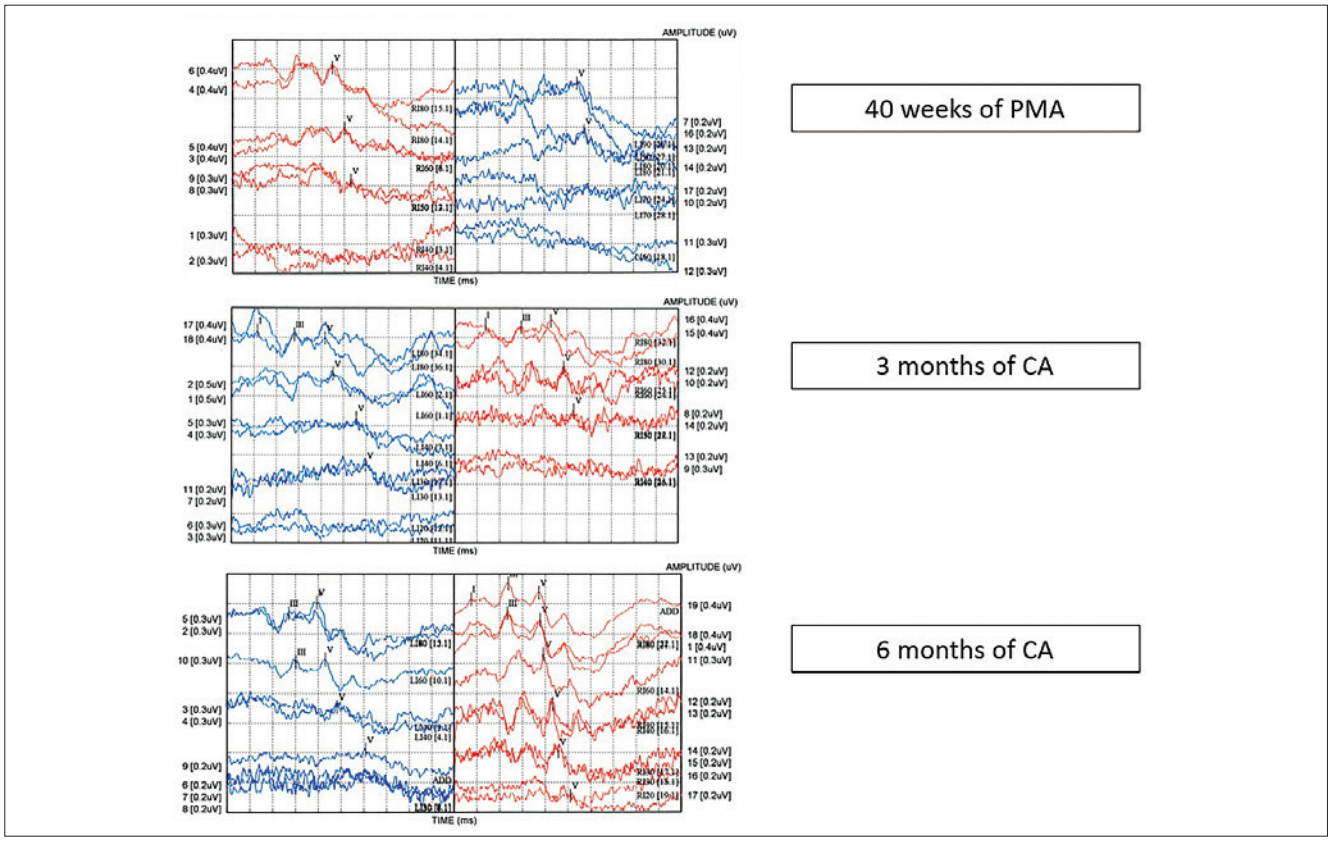

Fig. 3. Changes in hearing threshold in a preterm infant (GA 28.5 wk., PN 1160 g) in ABR performed at 40 weeks of postmenstrual age (PMA), 3 months and 6 months of corrected age (CA). The red traces refer to the stimulation of the right ear, the blue traces refer to the left one.

mean age of 6.3 (+ 1.3; range 5-8) months of CA for mild loss and of 8.6 (+ 3.4; range 5-12) months of CA for moderate loss.

Among infants with initial diagnosis of hearing loss, the threshold remained unchanged in 17 cases (53.1%): 10 cases of mild-moderate loss and all 7 cases (21.9%) with higher degree of bilateral loss (4 severe, 3 profound).

Among 17 patients affected by confirmed hearing loss, 3 infants showed unilateral (mild to moderate) loss. In these cases, audiological and phoniatric monitoring was proposed and later improvement with recovery to normal hearing was observed within 15 months of CA. In the remaining 14 cases affected by bilateral SNHL (7 moderate, 4 severe, 3 profound), acoustic amplification by hearing aids was indicated.

A definitive diagnosis of SNHL was reached by the fifth month of CA. When indicated, hearing aids were fitted at the mean age of 5.9 (± 1.3) months of CA.

None of our children were submitted to cochlear implantation (CI) during the time period of our study.

Table I shows the main neonatal features of infants with indication to aural habilitation (confirmed SNHL; 14 cases) and infants in whom amplification was not needed (normal and unilateral hearing loss; 18 cases). Infants addressed to amplification showed significantly lower GA and longer hospital stay in the NICU (GA 26.2 ± 2.2 weeks vs 28.4 ± 2.4; p = 0.0122 and NICU stay 132 ± 67 days vs 59 ± 27; p = 0.0002).

Discussion

Our study consists of an analysis of a homogeneous population of very preterm infants, admitted to "A. Gemelli" Hospital NICU, and followed prospectively after discharge. In our study we applied homogenous timing to perform initial electrophysiological evaluation by diagnostic ABR in all infants. We believe that this evaluation at 2-3 months allows for better comparison among infants and neonatal features, and reduces the possibility to detect temporary auditory dysfunction and, as a consequence, the risk of initial overestimation of hearing loss and an unjustified and dangerous increase in parental anxiety.

The prevalence of hearing loss in our group of very preterm infants was 13.4% at initial evaluation and 5.9% at confirmative diagnosis. This latter value is in substantial agreement with the available literature data [14 16].

We observed a trend towards improvement of hearing threshold in the first months of life in many infants, as already observed in several previous studies. In particular, 47% of our babies showed normalisation of initial mild-moderate hearing losses at final diagnosis. Coenraad et al. [1] showed an overestimation of hearing loss and an improvement in threshold (even with normalisation), in a heterogeneous population, also including term infants (GA median 34.7, interquartile range 27.3-39.3 weeks). Kang et al. [2] reported an improvement in hearing threshold even if these authors evaluated only 13 preterm subjects for 6 months, without providing any neonatal features. Preterm infants were also studied by Jiang et al. [17] who studied changes in hearing threshold in neonates born below 30 weeks of gestation at 28-42 of CA and found average changes from 28 dB at 28 weeks of CA to 13 dB at 42 weeks, without extending the audiological follow-up. Bovo et al. [11] studied 7 premature children within an heterogeneous group of potential candidates to CI and found improvement from a severe/profound degree of hearing loss up to mild degree/normalisation in all cases.

Hof et al. [9] retrospectively studied 14 preterm infants categorised as "refer" at hearing screening. These authors observed a tendency towards improvement of abnormal threshold in some subjects born at 28 weeks of GA or less. Among 3 children showing hearing improvement, two had moderate hearing loss with an initial diagnosis at one month of CA, while the third one had profound hearing loss initially diagnosed at only 33 weeks of PMA. Hof et al. [9] suggested that lower GA at birth and lower birth weight may be associated with better prognosis for hearing improvement.

After separating our infants in two subgroups, with and without indication for amplification, we found that infants with indication for hearing aids showed significantly lower GA and longer stay in NICU in comparison with infants without indication to audiological treatment. Taking into account the results of audiological follow-up, our data suggests that higher GA and shorter NICU stay represent favourable prognostic factors for hearing improvement within the first year of life. On the contrary, lower GA at birth and longer stay in NICU should be considered unfavorable prognostic factors for degree of hearing loss and improvement of hearing after initial electrophysiological diagnosis. Our results, based on a larger group of infants and with a homogeneous benchmark for initial diagnosis, is in contrast with those of Hof et al. [9].

Very premature infants are at high risk for hearing loss and their evaluation has to be prompt and accurate to ensure early (within the accepted boundary of 4-6 months of CA) activation of habilitation which can, in addition, promote auditory pathway maturation [18-20]. However, this category of children requires particular caution because of the commonly observed changes in auditory dysfunction. This aspect is critical in choice and timing of treatment, but also in communication and counseling to the parents of our patients. Accurate audiological surveillance is needed beyond the time of confirmative diagnosis. We found that most hearing threshold changes occur within about 6 months, but can be observed up to 12 months of CA. Our data suggest that changes in hearing threshold from initial diagnosis are more common in premature infants with higher GA at birth and shorter NICU stay and in mild/moderate degree of loss. On the contrary, our results suggest that changes are less likely to be found in infants with lower CA at birth and longer NICU stay and in cases with initial diagnosis of severe/profound hearing loss. However, we believe that reported observations of improvement (even to normalisation) of hearing in these categories of infants justify the recommendation of accurate audiological follow-up until 80-85 weeks of GA before changing habilitative indication, mostly in the case of CI [9 11 21 22].

Conclusions

Our study confirms the key role of audiological intervention in the management of very premature infants due to the prevalence and characteristics of hearing dysfunction in these patients.

We recommend initial electrophysiological evaluation of these infants performed at 2-3 three months of CA and followed by confirmative diagnosis within 5-6 months of CA.

Table I. Clinical characteristics of infants without hearing aids (normal or unilateral hearing loss) or with indication for hearing aid (confirmed bilateral SNHL).

	Infants without hearing aids (18 infants)	Infants with hearing aids (14 infants)	p
Gestational age (weeks - mean ± SD)	28.4 ± 2.4	26.2 ± 2.2	0.0122
Birth Weight (grams - mean ± SD)	1,045 ± 333	820 ± 330	ns
SGA (%)	22.2	28.6	ns
NICU stay (days - mean ± SD)	59 ± 27	132 ± 67	0.002

NICU: Neonatal Intensive Care Unit; SGA: Small for Gestational Age; SNHL: Sensorineural Nearing Loss; SD: Standard Deviation.

Accurate audiological follow-up in very premature infants should continue until 8-10 months of CA (in particular up to 80-85 weeks in candidates to CI).
Particular attention to possible changes in audiological diagnosis should be considered in infants with higher GA at birth, shorter admission in NICU and hearing loss of mild/moderate degree.
Further studies are needed to better delineate conditions associated with uncertainty and variations in audiological data in premature infants and to increase reliability of the procedures of audiological diagnostic set-up.

Conflict of interest statement

None declared.

References

1 Coenraad S, Goedegebure A, Hoeve J. *An initial overestimation of sensorineural hearing loss in NICU infants after failure on neonatal hearing screening.* Int J Pediatr Otorhinolaryngol 2011;75:159-62. https://doi.org/10.1016/j.ijporl.2010.10.026.

2 Kang MY, Jeong SW, Kim LS. *Changes in the hearing thresholds of infants who failed the newborn hearing screening test and in infants treated in the neonatal intensive care unit.* Clin Exp Otorhinolaryngol 2012;5(Suppl 1):S32-6. https://doi.org/10.3342/ceo.2012.5.S1.S32.

3 Colella-Santos MF, Hein TA, de Souza GL, et al. *Newborn hearing screening and early diagnostic in the NICU.* Biomed Res Int 2014;2014:845308. https://doi.org/10.1155/2014/845308.

4 American Academy of Pediatrics, Joint Committee on Infant Hearing. *Year 2007 position statement: principles and guidelines for early hearing detection and intervention programs.* Pediatrics 2007;120:898-921. https://doi.org/10.1542/peds.2007-2333.

5 Yoshinaga-Itano C. *Benefits of early intervention for children with hearing loss.* Otolaryngol Clin North Am 1999;32:1089-102.

6 Moeller MP. *Early intervention and language development in children who are deaf and hard of hearing.* Pediatrics 2000;106:E43. https://doi.org/10.1542/peds.106.3.e43.

7 Guzzetta F, Conti G, Mercuri E. *Auditory processing in infancy: do early abnormalities predict disorders of language and cognitive development?* Dev Med Child Neurol 2011;53:1085-90. https://doi.org/10.1111/j.1469-8749.2011.04084.x.

8 Lim HW, Kim EA, Chung JW. *Audiological follow-up results after newborn hearing screening program.* Clin Exp Otorhinolaryngol 2012;5:57-61. https://doi.org/10.3342/ceo.2012.5.2.57.

9 Hof JR, Stokroos RJ, Wix E, et al. *Auditory maturation in premature infants: a potential pitfall for early cochlear implantation.* Laryngoscope 2013;123:2013-8. https://doi.org/10.1002/lary.24054.

10 Jiang ZD, Wang J, Brosi DM, et al. *One-third of term babies after perinatal hypoxia-ischaemia have transient hearing impairment: dynamic change in hearing threshold during the neonatal period.* Acta Paediatr 2004;93:82-7.

11 Bovo R, Trevisi P, Ghiselli S, et al. *Is very early hearing assessment always reliable in selecting patiens for cochlear implants? A case series study.* Int J Pediatr Otorinolaringol 2015;79:725-31. https://doi.org/10.1016/j.ijporl.2015.02.033.

12 Martínez-Cruz CF, García Alonso-Themann P, Poblano A, et al. *Hearing loss, auditory neuropathy, and neurological co-morbidity in children with birthweight <750 g.* Arch Med Res 2012;43:457-63. https://doi.org/10.1016/j.arcmed.2012.08.007.

13 Borkoski-Barreiro SA, Falcón-González JC, Limiñana-Cañal JM, et al. *Evaluation of very low birth weight (≤ 1,500 g) as a risk indicator for sensorineural hearing loss.* Acta Otorrinolaringol Esp 2013;64:403-8. https://doi.org/10.1016/j.otorri.2013.05.002.

14 van Dommelen P, Verkerk PH, van Straaten HL; Dutch Neonatal Intensive Care Unit Neonatal Hearing Screening Working Group. *Hearing loss by week of gestation and birth weight in very preterm neonates.* J Pediatr 2015;166:840-3. https://doi.org/10.1016/j.jpeds.2014.12.041.

15 Engle WA; American Academy of Pediatrics Committee on Fetus and Newborn. *Age terminology during the perinatal period.* Pediatrics 2004;114:1362-4.

16 Robertson CM, Howarth TM, Bork DL, et al. *Permanent bilateral sensory and neural hearing loss of children after neonatal intensive care because of extreme prematurity: a thirty-year study.* Pediatrics 2009;123:e797-807. https://doi.org/10.1542/peds.2008-2531.

17 Jiang ZD, Yin R, Wilkinson AR. *Changes in hearing threshold between 28 and 42 weeks of age in babies born at under 30 weeks of gestation.* Acta Paediatr 2015;104:e143-7. https://doi.org/10.1111/apa.12920.

18 Kral A, O'Donoghue GM. *Profound deafness in childhood.* N Engl J Med 2010;363:1438-50. https://doi.org/10.1056/NEJMra0911225.

19 Sharma A, Dorman MF, Kral A. *The influence of a sensitive period on central auditory development in children with unilateral and bilateral cochlear implants.* Hear Res 2005;203:134-43. https://doi.org/10.1016/j.heares.2004.12.010.

20 Aimoni C, Crema L, Savini S, et al. *Hearing threshold estimation by auditory steady state responses (ASSR) in children.* Acta Otorhinolaryngol Ital 2018;38:361-8. https://doi.org/10.14639/0392-100X-1463.

21 Bovo R, Trevisi P, Zanoletti E, et al. *New trends in rehabilitation of children with ENT disorders.* Acta Otorhinolaryngol Ital 2017;37:355-67. https://doi.org/10.14639/0392-100X-1426.

22 Palma S, Roversi MF, Bettini M, et al. *Hearing loss in children with congenital cytomegalovirus infection: an 11-year retrospective study based on laboratory database of a tertiary paediatric hospital.* Acta Otorhinolaryngol Ital 2019;39:40-5. https://doi.org/10.14639/0392-100X-2020.

Received: February 15, 2018 - Accepted: October 7, 2018

How to cite this article: Frezza S, Catenazzi P, Gallus R, et al. *Hearing loss in very preterm infants: should we wait or treat?* Acta Otorhinolaryngol Ital 2019;39:257-262. https://doi.org/10.14639/0392-100X-2116

Address for correspondence: Piero Catenazzi, Department of Woman and Child Health, Obstetrics and Neonatology Area, Università Cattolica del Sacro Cuore, Fondazione Policlinico Universitario A. Gemelli IRCCS, largo F. Vito 1, 00168 Rome, Italy. Tel. +39 06 30154357. Fax +39 06 63055301. E-mail: piero.catenazzi@policlinicogemelli.it

ACTA OTORHINOLARYNGOLOGICA ITALICA 2019;39:263-268; doi: 10.14639/0392-100X-2190

VESTIBOLOGY

Height intolerance between physiological mechanisms and psychological distress: a review of literature and our experience

Mal d'altezza tra cause fisiologiche e stress psicologico: revisione della letteratura ed esperienza personale

R. TEGGI[1], F. COMACCHIO[2], F. FORNASARI[1], E. MIRA[3, 4]

[1] Department of Otolaryngology, Vita-Salute University, San Raffaele Hospital, Milan, Italy; [2] Otolaryngology and Otosurgery Unit, University Hospital of Padova, Italy; [3] Department of Otolaryngology, University of Pavia, Italy; [4] Department of Otolaryngology, Policlinico S. Matteo, Pavia, Italy

SUMMARY

Height intolerance often includes various clinical conditions ranging from physiological height instability, which is a common condition, to acrophobia, considered to be a specific phobia in the *Diagnostic and Statistical Manual of Mental Disorders*, 5th edition (DSM-V). Visual dependence is commonly reported in patients with height intolerance, and physiological mechanisms may include conflicting information from visual cues on one hand and vestibular-proprioceptive cues on the other. This study examines the physiological mechanisms underlying height intolerance and phobic-cognitive mechanisms leading to more severe clinical manifestations (i.e. acrophobia). Diagnosis mainly relies on history, while the use of specific questionnaires has been proposed in a clinical setting. Treatment includes physical therapy with the purpose of habituation to the condition; on the other hand, psychological disorders should be considered and treated. Finally, our own experience in treating patients with height intolerance is included. In a sample of 164 acrophobic patients with imbalance lasting for at least 6 months, a prevalence of females was found (59.7%); among comorbidities, motion sickness (51.8%), migraine (50.6%) and panic disorders (18.9%) were reported. Interestingly, acrophobia always preceded the first panic attack.

KEY WORDS: Fear of height • Height intolerance • Acrophobia • Vestibular system • Phobic disorders

RIASSUNTO

Il termine "mal d'altezza" comprende spesso condizioni cliniche differenti che vanno dalla fisiologica instabilità, che può essere considerata una condizione comune, all'acrofobia, che è stata inclusa tra le forme fobiche specifiche nella quinta edizione del Manuale Diagnostico e Statistico dei Disturbi Mentali (DSM-V). Una dipendenza dalle informazioni visive è comunemente descritta nei pazienti con mal d'altezza; i meccanismi fisiologici alla base del disturbo potrebbero essere le informazioni contrastanti provenienti dal sistema visivo da un lato e propriocettivo-vestibolare dall'altro. Il nostro studio esamina le pubblicazioni relative ai meccanismi fisiologici che producono il mal d'altezza e ai disturbi di tipo fobico-cognitivo che producono le manifestazioni cliniche più severe (i.e. acrofobia). La diagnosi si basa essenzialmente sulla storia clinica e l'uso di questionari specifici è stato proposto come il migliore metodo di indagine. Il trattamento comprende una fisioterapia specifica con lo scopo di abituare il paziente alla condizione; d'altro canto i disturbi psicologici che producono i quadri clinici più severi devono essere indagati e trattati. Infine, abbiamo incluso la nostra esperienza clinica sull'argomento. In un campione di 164 pazienti acrofobici e con instabilità presente da almeno 6 mesi, abbiamo trovato una prevalenza del sesso femminile (59,7%); tra le comorbidità la cinetosi (51,8%), l'emicrania (50,6%) e le patologie da panico (18,9%) erano i più comuni. Può essere rilevante notare come in questi soggetti l'acrofobia abbia sempre preceduto il primo attacco di panico.

PAROLE CHIAVE: *Vertigine da altezza • Intolleranza all'altezza • Acrofobia • Sistema vestibolare • Disordini fobici*

Introduction

Height vertigo or visual intolerance to height refers to psychological, neurovegetative and behavioural disorders that affect predisposed individuals following exposure to height. It is the core part of a spectrum of manifestations ranging from physiological instability to height and acrophobia [1] The predisposing factor is thought to be conflicting information arising from vestibular, somatosensory and visual cues when there is an excessive distance of the stationary reference frame at the periphery of the visual field preventing perception of the body's oscillations, which is required for locomotion and to maintain

an upright position; psychological factors may also play a role in the disorder [2]. It affects about a third of the general population, and in about half of cases significantly impacts the quality of life. It can be treated with appropriate recommendations or with proper rehabilitation, psychotherapy and drugs [3].
The research was performed on PUBMED and SCOPUS. To clarify our selection criteria a flowchart is shown (Fig. 1).

A proposed physiological mechanism for height intolerance

Correct postural control and correct spatial orientation are essential requirements for human survival and are the result of an evolutionary pathway that started around 6 million years ago when we gained the upright position. It requires univocal information from vestibular, somatosensory (tactile and proprioceptive) and visual cues. The tactile component is mainly represented by pressure sensors on the sole of the foot, providing information on how the foot approaches the ground, while the proprioceptive component, represented by the musculotendinous and articular receptors, informs us on the relationships between the different parts of our body (trunk, neck, limbs). The vestibular system works as a sensor for head accelerations and, more importantly, as a sensor for gravity. The visual system, both foveal and peripheral, gives us information about the external environment: the shape, size, distance and movements of the objects around us and movements of our head in the visual scene [1].
When we stand still in a static environment, our postural control is maintained through continuous small oscillations laterally and back to front. In this situation, visual information overcomes somatosensory and vestibular information, reducing oscillations by 50-100% [4].
In order to detect the displacement of an object by the eye, the image must slide on the peripheral retina by at least 20' of arc. Knowing that we oscillate about 2 cm laterally under normal conditions, the 20' of arc threshold is reached in a static field of view at a distance less than 3 m. If this distance increases, the amplitude of the retinal slip is reduced with resulting conflicting information between the somatosensory and vestibular systems on the one hand, perceiving our oscillations, and the visual system on the other, perceiving a static visual field [3-7]. This is what physiologically happens when there are no static reference points near our field of vision, as on a mountain or on a roof or on a terrace of a tall building [2]. This conflicting situation can be avoided or reduced by increasing the amplitude of postural sways in order to increase visual control. It has been calculated that the amplitude of sways of our body cannot exceed 10 cm, corresponding to a height of 20 m, which represents the threshold beyond which one is not able to maintain an upright position and increases the risk of falls [3].

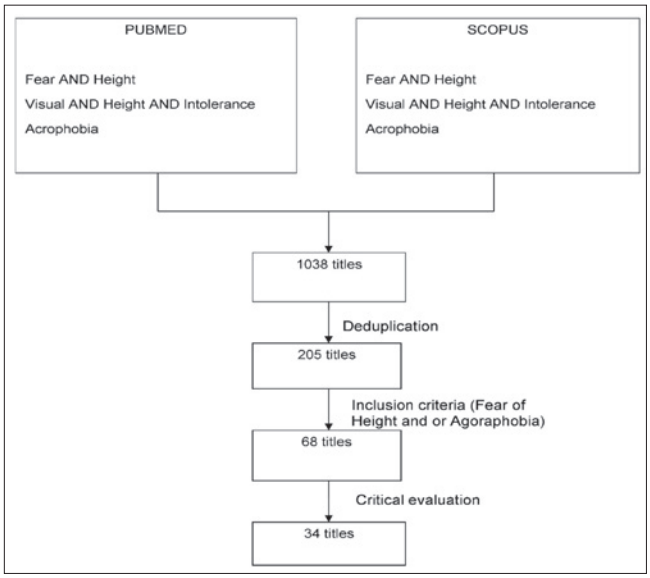

Fig. 1. Search strategy flowchart.

The reduction or loss of visual information to postural control in all individuals (100%) provokes a variable amount of instability that can be considered physiological and defined as *physiological visual instability at height*. In susceptible individuals (around 30% of the general population), the same condition generates a state of discomfort and anxiety with neurovegetative and motor symptoms. This condition is considered to be pathological and is called *height vertigo or visual intolerance to height*. Finally, in 6.4% of the general population (8.6% of women vs 4.1% of men), this condition is so unpleasant that it becomes a genuine phobia, as defined according to the criteria of the *Diagnostic and Statistical Manual of Mental Disorders*, 5th edition (DSM-V), requiring treatment: *acrophobia or fear of height* [8-10].
On the other hand, some individuals, known as risk takers/risk seekers, take pleasure from the feeling of danger linked to exposure to height and the resultant release of catecholamines, and look for it voluntarily [11].

Epidemiology

Height intolerance affects 28.5% of the general population; it is more common in women (32.4%) than in men (24.5%); it can begin at any age, but manifests more frequently (30%) in the second decade of life with the highest prevalence being observed in women during the fifth decade [5]. It often has a familiar trait and is correlated with migraine, motion sickness, anxiety disorders and depression [12].

After initial onset, it can spontaneously improve in 31% of cases, persist unchanged in 52% and worsen in 13% [5]. In about half of cases, it becomes clinically significant and considerably impacts the quality of life. In more than 20% of cases, it can cause panic attacks and in 6.4% of cases, it worsens significantly to become acrophobia [5-7]. A recent study focused on general phobic aspects of height intolerance using the General Self-Efficacy Scale, a 10-item psychometric scale designed to assess optimistic self-beliefs to cope with a variety of difficult demands in life. In it, the authors demonstrated an association between this scale and height intolerance [13]. Primary school children can be affected by height intolerance, although it usually remits within a few years, possibly facilitated by repeated exposure to the triggering situations [14]. An overlap with agoraphobia has been supposed, possibly based on commonly shared pathophysiological mechanisms (i.e. increased visual dependence in postural control) [15]. Specific phobias are more frequently associated with higher alcohol consumption; on the other hand, a recent questionnaire-based study did not provide evidence of an association in subjects with height intolerance [16].

Clinical features: symptoms, signs and diagnosis

Looking down from a tower or from the roof or from the balcony of a tall building, climbing or descending a ladder, walking on a bridge or on a mountain, climbing, walking on a ridge or on an exposed path, travelling on a cable car or a chairlift are all conditions that can cause height vertigo [5].
The intensity of the disorder is strongly correlated to body position: it is highest when standing, it decreases when sitting or bending the knees and becomes minimal or disappears when lying down in the prone position. Vestibular, neurological or visual disorders reducing visual acuity from afar can increase the disorder [3]. It is more likely to manifest in some environmental situations that interfere with vision such as fog or semi-darkness, with some tactile and proprioceptive signals such as slippery ground, and with specific vestibular stimulations such as hyperextension of the head, which moves the macular receptors into an unnatural position. On the other hand, the somatosensory afferents that arise from leaning against a wall with your back or your side, the tactile stimulation of the hand touching a banister or a firm solid structure can relieve it. The same applies to the presence of nearby stationary reference points that are visible on the periphery of our visual field. In any case, the psychological attitude of the subject plays a lead role in the genesis of visual intolerance to height because the level of anxiety and fear of the void can overestimate height perception when looking from above [17].
The disorders arise in predisposed individuals from the interplay of postural and eye-gaze changes in a threatening situation with psychological factors. In fact, physiological postural reactions may be recorded in healthy subjects when exposed to height even if virtual [18]. According to some authors, subjects with a fear of heights are probably more prone to rely on visual information in postural control, compared to normal subjects. When exposed to optokinetic visual stimuli, they exhibit higher anxiety and an increased body sway [19]. Subjects with height vertigo present an alteration of the standing position and gait, and of head and eye movements that are essential in exploring the surroundings. In an experimental setting, it has been demonstrated that exposure to height provokes changes in postural control, mainly increasing co-contraction of leg muscles; moreover, the severity of these changes correlates with anxiety [20]. Similarly, subjects with height intolerance walked more slowly on a 15 metre balcony, with reduced cadence and stride length; no changes were recorded when subjects performed the test walking with upward gaze or with eyes closed [21].
Likewise, changes in visual exploration have been demonstrated during height exposure. Individuals with height intolerance exhibit fewer and smaller amplitude eye-in-head saccades with a longer fixation time than in less susceptible subjects. Moreover, spontaneous head movements were reduced with all three dimensions equally affected. Gaze-in-space, which expresses the ability to explore the surroundings by coordinated eye-head movements, covered a smaller total area of the visual scene [8 22 23]. A recent paper showed that the height condition affects optokinetic gain, which was increased, and smooth pursuit, supporting neuro-anatomical evidence of threat-related mechanisms influencing both oculomotor nuclei and vestibular reflex pathways; the authors emphasise how anxiety and cognitive activity may play a role in the performance of eye movements [24]. Their results agree with the findings of previous studies, underlining the interference of fear on postural control and eye movements [11 25].
It could be argued that, as a consequence of these changes, subjects with height intolerance exhibit a more cautious, slow and rigid gait with reduced speed and length of steps when exposed to the triggering condition; saccades are reduced in number and amplitude, they last longer and are mainly directed on the horizontal plane. Spontaneous movements of the head are greatly reduced in number and speed in all directions and, consequently, the gaze-in-space, which represents an indicator of eye-head movement coordination, covers a smaller area of the visual

field and is strictly directed (*frozen*) toward the horizon. Changes of gait and eye movements are often associated with distress and neurovegetative symptoms [26].

Since there is no conclusive test to diagnose height intolerance and its impact on quality of life, clinical history and above all, questionnaires are, at present, the best choice for this purpose. For evaluation of symptoms, they must be predictive of subjective dizziness, psychological distress and avoidance [27 28].

Treatment

When height intolerance becomes more severe and presents a considerable impact on quality of life, it can be included among phobias (acrophobia) based on DSM-V criteria [10] and must be treated accordingly with pharmacotherapy, psychotherapy and behavioural therapy.

Fortunately, the most common presentation of visual intolerance to height is of little clinical relevance and mostly requires suggestions and recommendations, such as avoiding exposure to heights and certain sports like climbing or mountaineering, to manage both the neurophysiological and psychological aspects of the disturbance. However, if a susceptible individual is exposed to height, some practical advice might be sufficient, such as to stop walking, to sit or lie down, to avoid looking down or far away but to fix on stable and nearby structures or to close the eyes, to reduce movement or hyperextension of the head, to lean against a fixed support even if only with the hand, to avoid wearing multifocal glasses or ski goggles, which prevent lateral peripheral vision, and to be mentally engaged in some cognitive tasks to shift attention from the apprehension of a possible fall [8 11].

Only more complex cases need rehabilitative therapy. Based on the visual dependence of these subjects, some authors have proposed exercise protocols not so different from those used in patients with agoraphobia and based on habituation; the phobic component must also be considered and treated with cognitive behavioural therapy [29-32].

Recent papers have focused on the possibility of using virtual reality in the habituation techniques, as well as using hypnosis, and these might offer an opportunity to decrease the phobic-cognitive disorders of more complex cases [33 34].

Conclusions

Height vertigo or visual intolerance to height is a very common syndrome that manifests in susceptible individuals following exposure to height that has been described since ancient times. Its relationship with vestibular disorders, in particular with persistent postural perceptual dizziness (PPPD), is still matter of debate.[35,36] It mainly presents with fear of falling or losing equilibrium, neurovegetative symptoms, reduced visual exploration and generalised contraction of antigravity muscles with a rigid and cautious attitude to standing position and gait. It originates from the interaction between psychological factors, mainly anxiety, and organic factors, such as the intersensorial conflict between visual, vestibular and somatosensory systems involved in postural control. It lies in the middle of a spectrum of disorders related to height exposure ranging from physiological visual instability to height up to acrophobia or fear of heights. Further research will allow a better understanding of the mechanisms behind this complex disorder that strongly impacts on daily activities, on interpersonal relationships, and on overall quality of life.

Our experience

In order to assess comorbidities and clinical vestibular signs, among the records of 4850 outpatients who attended the tertiary Centre for Vestibular Disorders of San Raffaele Hospital between 2006 and 2017, we found 164 subjects referring fear of height and chronic dizziness without a lifetime history of vertigo of any kind. The inclusion criterion was the presence of both disorders for at least 6 months before consultation. Subjects were included if they experienced fear of height interfering with the activities of everyday life, such as standing on a ladder or a chair; moreover, they should have referred a persistent sensation of rocking or swaying, unsteadiness and/or dizziness without vertigo that had been present at least for the last 6 months. The age of onset of both disorders was noted. The mean age of the sample was 41.5 ± 8.7 years. Ninety-eight (59.7%) were female. A full clinical history was collected before examination, in particular for motion sickness, lifetime history of migraine and panic disorders (PD). Clinical examination included otoscopy, audiometric exam, Head Impulse test (from November 2013, a video HIT was performed), full bedside examination with video Frenzel, including positional tests, head shaking test and a 100 Hz vibratory test. Moreover, a static stabilometric exam (S.Ve.P - Amplaid) was performed. Results were compared with those of 100 normal subjects chosen to overlap for age and sex with controls (mean age 42 ± 7, 60 females). The severity of dizziness was measured by the 25-item Dizziness Handicap Inventory scale, validated Italian version, which generated a total score (range zero to 100) indicating the patient's self-perceived level of handicap associated with the dizziness [37 38]. DHI was further subdivided into physical (ranging from 0 to 28 points), functional (ranging

Table I. Values of stabilometric parameters Length (L) and Surface (S) in the two groups in the tested conditions: eyes open (eo) and eyes closed (ec). Values are expressed as mean ± standard deviation. Length is expressed in millimeters while Surface in squared millimeters. A quotient has been calculated for the value of L_{eo}/L_{ec}.

	Controls (n = 100)	Patients (n = 164)	Statistical analyses
Length eyes open	195 ± 36	202 ± 58	n.s.
Length eyes closed	302 ± 44	449 ± 65	.0001
Surface eyes open	186 ± 34	198 ± 39	.01
Surface eyes closed	239 ± 47	376 ± 55	.0001
Q (L_{eo}/L_{ec})	64 ± 5	45 ± 4	.0001

between 0 and 36 points) and emotional (ranging from 0 to 36 points) subscores. A higher score indicates a more severe handicap.

Among comorbidities, lifetime episodes of headaches with migrainous features (pulsatile, associated with phonophobia and photophobia, lasting for more than 4 hours and worsening on exertion) were reported by 83 of 164 (50.6%) subjects; it was more frequent in females, since 59 of 98 females (60.2%) reported it, whereas 24 of 66 males (36.4%) reported it. Previous episodes of panic attacks requiring therapy were reported by 31 subjects (18.9%), with no difference between the two sexes, since 19 of 98 females (19.4%) and 12 of 66 males (18.2%) referred it. An association was found between migraine and panic disorders (PD) since among 83 migraineurs, 28 reported panic disorders whereas only 3 of 81 non-migraineurs reported them ($\chi^2 = 12.9$, $p = 0.0003$). Among 31 PD subjects, fear of height preceded the first panic attacks in all subjects.

Eighty-five (51.8%) subjects reported motion sickness as children, while 59 (36.0%) still suffered from it as adults. Finally, vestibular bedside examination was negative in 132 subjects. A long-lasting smooth bipositional apogeotropic or geotropic nystagmus was found in 18 subjects (11.0%), a positive skull vibration test in 12 subjects (7.1%) and a positive head shaking test in 2 patients (1.2%); 3 subjects presented both a bipositional nystagmus and a vibration-induced nystagmus.

Results of static stabilometry are summarised in Table I. Stabilometric findings demonstrated increased values of parameters in patients above all in eyes closed conditions, underlining an increased dependence on visual cues. Results of the DHI questionnaire are reported in Table II.

The DHI comprises 25 items; with a total score ranging between 0 and 100 points.

Although our results are far from being conclusive, comparing the prevalence of comorbidities with previous studies on the general population, some conclusions can be drawn. Above all in our sample, the frequency of migraineurs (50.6%) was higher than in a previous study,

Table II. Values of the Dizziness Handicap Inventory total score and subscales in patients (n = 164).

DHI total score	DHI subscales		
	Emotional	Physical	Functional
24.7 ± 9.4	6.4 ± 2.6	6.3 ± 2.6	11.9 ± 4.8

which reported a prevalence of 18.2% among females and 6.5% among males [39]. In the same way, PD was most represented among our subjects than in the general population, in which a previous study estimated the prevalence to be in a range from 1.4% to 2.9% [40]. On the other hand, it should be considered that a possible bias may arise from our inclusion criteria, since our subjects presented fear of height interfering with their daily activities and also reported chronic dizziness, so it can be argued that they presented acrophobia in a burden of symptoms of PPPD.

Conflict of interest statement

None declared.

References

1. Bronstein AM, Brandt T, Woollacott M. *Clinical disorders of balance, posture and gait*. London: Arnold; 1996.
2. Brandt T, Bles W, Arnold F, et al. *Height vertigo and human posture*. Adv Otorhinolaryngol 1979;25:88-92.
3. Brandt T, Arnold F, Bles W, et al. *The mechanism of physiological height vertigo. I. Theoretical approach and psychophysics.* Acta Otolaryngol 1980;89:513-23. https://doi.org/10.3109/00016488009127169.
4. Bles W, Kapetyn TS, Brandt T, et al. *The mechanism of physiological height vertigo. II. Posturography.* Acta Otolaryngol 1980;89:534-40. https://doi.org/10.3109/00016488009127171.
5. Huppert D, Grill E, Brandt T. *Down on heights? One in three has visual height intolerance*. J Neurol 2013;260:597-604. https://doi.org/10.1007/s00415-012-6685-1.
6. Brandt T, Huppert D. *Fear of heights and visual height intolerance*. Curr Opin Neurol 2014;27:111-7. https://doi.org/10.1097/WCO.0000000000000057.
7. Nakahara H, Takemori S, Tsuruoka H. *Influence of height on the spatial orientation and equilibrium of the body*. Otolaryngol Head Neck Surg 2000;123:501-4. https://doi.org/10.1067/mhn.2000.107316.
8. Brandt T, Kugler G, Schniepp R, et al. *Acrophobia impairs visual*

exploration and balance during standing and walking. Ann N Y Acad Sci 2015;1343:37-48. https://doi.org/10.1111/nyas.12692.

9 Brandt T, Benson J, Huppert D. *What to call "non phobic" fear of heights?* Br J Psychiatry 2012;190:81. https://doi.org/10.1192/bjp.190.1.81a.

10 American Psychiatric Association. *Diagnostic and statistical manual of mental disorders*. *DSM-5*. Fifth Edition. Washington, DC: American Psychiatric Association Publishing; 2013.

11 Salassa JR, Zapala DA. *Love and fear of heights: the pathophysiology and psychology of height imbalance*. Wilderness Environ Med 2009;20:378-82. https://doi.org/10.1580/1080-6032-020.004.0378.

12 Kapfhammer HP, Huppert D, Grill E, et al. *Visual height intolerance and acrophobia: clinical characteristics and comorbidity patterns*. Eur Arch Psychiatry Clin Neurosci 2015;265:375-85. https://doi.org/10.1007/s00406-014-0548-y.

13 Grill E, Schäffler F, Huppert D, et al. *Self-efficacy beliefs are associated with visual height intolerance: a cross-sectional survey*. PLoS One 2014;9:e116220. https://doi.org/10.1371/journal.pone.0116220.

14 Huppert D. *[Fear of heights in primary school children]*. Fortschr Neurol Psychiatr 2016;84:134-6. https://doi.org/10.1055/s-0042-103424.

15 Davey GC, Menzies R, Gallardo B. *Height phobias and biases in the interpretation of bodily sensations: some links between acrophobia and agoraphobia*. Behav Res Ther 1997;35:997-1001.

16 Huppert D, Grill E, Kapfhammer HP, et al. *Fear of height and mild visual height intolerance independent to alcohol consumption*. Brain Behav 2013;3:596-601. https://doi.org/10.1002/brb3.162.

17 Stefanucci, JK, Proffitt DR. *The roles of altitude and fear in the perception of height*. J Exp Psychol Hum Percept Perform 2009;35:424-38. https://doi.org/10.1037/a0013894.

18 Cleworth RW, Chua R, Inglis TJ, et al. *Influence of virtual height exposure on postural reactions to support surface translations*. Gait Posture 2016;47:96-102. https://doi.org/10.1016/j.gaitpost.2016.04.006.

19 Hüweler R, Kandil FI, Alpers GW, et al. *The impact of visual flow stimulation on anxiety, dizziness and body sway in individuals with and without fear of heights*. Behav Res Ther 2009;47:345-52. https://doi.org/10.1016/j.brat.2009.01.011.

20 Wuehr M, Kugler G, Schniepp R, et al. *Balance control and antigravity muscle activity during the experience of fear at heights*. Physiol Rep 2014;2:e00232. https://doi.org/10.1002/phy2.232.

21 Schniepp R, Kugler G, Wuehr M, et al. *Quantification of gait changes in subjects with visual height intolerance when exposed to heights*. Front Hum Neurosci 2014;8:963. https://doi.org/10.3389/fnhum.2014.00963.

22 Kugler G, Huppert D, Schneider E, et al. *Fear of heights freezes gaze to the horizon*. J Vestib Res 2014;24:433-41. https://doi.org/10.3233/VES-140529.

23 Kugler G, Huppert D, Eckl M, et al. *Visual exploration during locomotion limited by fear of heights*. PLoS One 2014;9:e105906. https://doi.org/10.1371/journal.pone.0105906.

24 Naranjo EN, Cleworth TW, Allum JHJ, et al. *Threat effect on human oculo-motor function*. Neuroscience 2017;359:289-98. https://doi.org/10.1016/j.neuroscience.2017.07.024.

25 Boffino CC, de Sá CS, Gorenstein C, et al. *Fear of heights: cognitive performance and postural control*. Eur Arch Psychiatry Clin Neurosci 2009;259:114-9. https://doi.org/10.1007/s00406-008-0843-6.

26 Coelho CM, Wallis G. *Deconstructing acrophobia, physiological and psychological precursors to developing a fear of height*. Depress Anxiety 2010;27:864-70. https://doi.org/10.1002/da.20698.

27 Steinman SA, Teachman BA. *Cognitive processing and acrophobia: validating the heights interpretation questionnaire*. J Anxiety Disord 2011;25:896-902. https://doi.org/10.1016/j.janxdis.2011.05.001.

28 Huppert D, Grill E, Brandt T. *A new questionnaire for estimating the severity of visual height intolerance and acrophobia by a metric interval scale*. Front Neurol 2017;8:211. https://doi.org/10.3389/fneur.2017.00211.

29 Arroll B, Henwood SM, Sundrama FI, et al. *A brief treatment for fear of heights: A randomized controlled trial of a novel imaginal intervention*. Int J Psych Med 2017;52:21-33. https://doi.org/10.1177/0091217417703285.

30 Arroll B, Wallace HB, Mount V, et al. *A systematic review and meta-analysis of treatments for acrophobia*. Med J Aust 2017;206:263-7.

31 Whitney SL, Jacob RG, Sparto PJ, et al. *Acrophobia and pathological height vertigo: indications for vestibular physical therapy?* Phys Ther 2005;85:443-58.

32 Dreyer-Oren SE, Clerkin EM, Edwards CB, et al. *Believing is seeing: changes in visual perception following treatment for height fear*. J Behav Ther Exp Psychiatry 2019;62:1-6. https://doi.org/10.1016/j.jbtep.2018.08.001.

33 Coelho CM, Walters AM, Hine TJ, et al. *The use of virtual reality in acrophobia research and treatment*. J Anxiety Disord 2009;23:563-74. https://doi.org/10.1016/j.janxdis.2009.01.014.

34 Hirsch JA. *Integrating hypnosis with other therapies for treating specific phobias: a case series*. Am J Clin Hypn 2018;60:367-77. https://doi.org/10.1080/00029157.2017.1326372.

35 Bisdorff A, von Brevern M, Lempert T ,et al. *Classification of vestibular symptoms: Towards an international classification of vestibular disorders*. J Vestib Res 2009;19:1-13. https://doi.org/10.3233/VES-2009-0343.

36 Staab JP, Eckhardt-Henn A, Horii A, et al. *Diagnostic criteria for persistent postural-perceptual dizziness (PPPD): Consensus document of the committee for the Classification of Vestibular Disorders of the Barany Society*. J Vestib Res 2017;27:191-208. https://doi.org/10.3233/VES-170622.

37 Jacobson GP, Newman CW. *The development of the Dizziness Handicap Inventory*. Arch Otolaryngol Head Neck Surg 1990;116:424-7.

38 Nola G, Mostardini C, Salvi C, et al. *Validity of Italian adaptation of the Dizziness Handicap Inventory (DHI) and evaluation of the quality of life in patients with acute dizziness*. Acta Otorhinolaryngol Ital 2010;30:190.

39 Lipton RB, Stewart WF, Diamond S, et al. *Prevalence and burden of migraine in the United States: data from the American Migraine Study II*. Headache 2001;41:646-57.

40. Weissman MM, Bland RC, Canino GJ, et al. *The cross-national epidemiology of panic disorder*. Arch Gen Psychiatry 1997;54:305-9. https://doi.org/10.1001/archpsyc.1997.01830160021003.

Received: May 5, 2018 - Accepted: October 6, 2018

How to cite this article: Teggi R, Comacchio F, Fornasari F, et al. *Height intolerance between physiological mechanisms and psychological distress: a review of literature and our experience.* Acta Otorhinolaryngol Ital 2019;39:263-268. https://doi.org/10.14639/0392-100X-2190

Address for correspondence: Roberto Teggi, San Raffaele Hospital, via Olgettina 60, 20132 Milan, Italy. E-mail: teggi.roberto@hsr.it

ACTA OTORHINOLARYNGOLOGICA ITALICA 2019;39:269-273; doi: 10.14639/0392-100X-2309

Otology

3T MRI-based estimation of scalar cochlear implant electrode position

Valutazione con RMN 3-T della posizione degli elettrodi degli impianti cocleari nella scala cocleare

F. TEK[1], S. MÜLLER[1], E. BOGA[2], H.B. GEHL[2], D. SEITZ[1], L.U. SCHOLTZ[1], H. SUDHOFF[1], I. TODT[1]

[1] Department of Otolaryngology, Head and Neck Surgery, [2] Department of Radiology
Klinikum Bielefeld, Ruhr Universität Bochum, Germany

SUMMARY

Common techniques to evaluate intracochlear electrode position include ionised radiation by multi-slice computer tomography, digital volume tomography (MSCT, DVT) and flat panel tomography (FPT). Recent advances in the knowledge about handling MRI artefacts and the pain-free performance of MRI scans in cochlear implantees showed that estimation of the intracochlear electrode position is possible at 1.5 T with perimodiolar or midmodiolar arrays. The aim of the present study is to evaluate the assessment of the ipsilateral scalar position of a cochlear implant lateral wall electrode by MRI sequences at 3T. In a prospective study we evaluated 10 patients implanted with a diametrically bipolar implant magnet system with a lateral wall electrode in the intrascalar electrode position in an axial and coronal position and a T2 weighted sequence at 3T and a resolution of 0.8 mm. We compared the intracochlear position with routine postoperative DVT scan. In all cases, the MRT-estimated scalar position corresponded with that estimated by DVT scan. In all cases, a scala tympani position was present. While the position in the basal turn is reliably localisable, the first-turn visual assessment is difficult. Estimation of the intracochlear position of lateral wall cochlear implant electrodes by 3T MRI is possible for the basal turn. Electrode design plays a major role in visual assessment.

KEY WORDS: MRI • Cochlear implant • Electrode position

RIASSUNTO

La valutazione del posizionamento dell'elettrodo di un impianto cocleare avviene generalmente mediante tecniche che utilizzano radiazioni ionizzanti come la tomografia computerizzata multi-slice (MSCT), la tomografia a volume digitale (DVT) o la tomografia flat-panel (FPT). Recenti sviluppi nella gestione degli artefatti in risonanza magnetica (MRI) e la possibilità di eseguire MRI senza arrecare comorbidità al paziente impiantato, hanno dimostrato che l'applicazione della MRI a 1,5 T è possibile per stimare il posizionamento dell'elettrodo di un impianto cocleare. Lo scopo dello studio è indagare il posizionamento scalare di un elettrodo a parete per mezzo di MRI a 3 T. In questo studio prospettico sono stati arruolati 10 pazienti sottoposti ad impianto cocleare dotato di magnete bipolare, con elettrodo posizionato a livello intrascalare a parete. I pazienti sono stati valutati con MRI a 3 T, in sequenze T2-pesate con risoluzione di 0,8 mm, assiali e coronali. La posizione intracocleare dell'elettrodo osservata nelle sequenze MRI è stata comparata con quella ottenuta in DVT, eseguita routinariamente nel periodo postoperatorio. In tutti i casi la posizione scalare dell'elettrodo stimata in MRI era sovrapponibile a quella evidenziata in DVT. In tutti i casi è l'elettrodo è stato osservato nella scala timpanica. La posizione dell'elettrodo è stata valutata in modo affidabile a livello del giro basale della coclea, al contrario, la visualizzazione dell'elettrodo nei giri cocleari successivi è risultata difficoltosa. La valutazione con tecniche di MRI a 3 T del posizionamento dell'elettrodo a parete è possibile per il giro basale della coclea. La tipologia di elettrodo gioca un ruolo fondamentale nella stima visiva.

PAROLE CHIAVE: *MRI • Impianto cocleare • Posizionamento dell'elettrodo*

Introduction

Estimation of intracochlear electrode position after CI electrode insertion is of high importance for audiological outcomes [1]. Radiological tools associated with ionised radiation (CT, DVT, FPT) are mainly used to clarify this important question for the surgeon, technician and audiologist. Recent observations show of the possibility to perform radiation-free positional estimations of electrode position. One option is positional estimation based on intraoperative electrophysiological measurements [2], with limitations in terms of the electrodes used and brand-specific intraoperative electrophysiological measurement abilities.

Another option is the use of MRI scans to clarify this

clinically important question. Due to the internal magnet, MRI scans can be associated with complications such as pain or magnet dislodgements [3 4] at 1.5T, and scans cannot be performed without removal of the magnet at 3T (current implant series, Cochlear Company, Sydney, Australia). This observation limits the utility of MRI scans.
New studies show that the specific positioning of the implant magnet allow MRI-based visual assessment of the internal auditory canal and the cochlea-even after the implantation is performed [5 6]. Recently, it was shown that estimation of the electrode position at 1.5T for perimodiolar or midmodiolar electrodes is possible [7].
3T scanning is known to provide increased visual resolution. Diametrically bipolar internal magnet systems containing CI systems offer the opportunity of 3T MRI scanning without complications [8]. This system contains an electrode that is positioned at the lateral wall of the cochlea. Lateral wall electrodes are known for their less effective visual localisation abilities in the CT, DVT and FTP in comparison to perimodiolar or midmodiolar electrodes, due to their generally higher lateral position in the scala tympani.
The aim of the present study was to evaluate assessment of the ipsilateral scalar position of cochlear implant lateral wall electrodes by MRI sequences at 3T.

Materials and Methods

The study was approved by the institutional review board of Klinikum Bielefeld, Germany (IRB-klibi-HNO-2017/05). Patients gave written informed consent for use of clinical records.
In this prospective study, 10 patients underwent 3T MRI scanning in a tertial referral centre. Between May 2017 and December 2017, all patients were implanted with a MED-EL SYNCHRONY implant (MED-EL, Innsbruck, Austria) with a diametrically magnetised internal magnet. In all cases, the implant magnet was intraoperatively determined and positioned 7-9 cm behind the external auditory canal.
All examinations were performed in a 3T MR imaging unit (Achieva, Philips Medical Systems, Best, NL) without a headband on the first postoperative day. Additionally, cone beam CT (NEW TOM VGI, Verona, Italy) was performed.
MRI scanning parameters:
TSE T2 2D: TR: 3000ms, TE 120ms, slice thickness 0.8 mm, voxel size 0.449 mm, F0V 230×199; 35 slices.
CBCT parameters:
FOV 15x15 cm, 10,48 mAS-20,52 mAS, KV 110, 360° followed by 2D and 3D reconstruction at an external workstation (NNT, main station).

Results

In all patients MRI scanning was performed without any pain or discomfort. Related to a scanning slice thickness of 0.8 mm, 7 to 8 pictures of the electrode inserted in the cochlea in the axial view were visible. In all scala tympani-positioned cases, visualisation of the basal turn was possible with a diminished signal of the scala tympani and a persistent signal in the scala vestibuli (Fig. 2a). During the first turn, the signal at the axial overview was visible with improved quality compared to a previous study [7]. The comparison between the regular axial view without an electrode (Fig. 1a,b) and the inserted electrode (Fig. 2b) allowed for estimation of the inserted electrode in the first turn. A diminishing signal, which allows a differentiation between a scala tympani or a scala vestibuli position of the electrode, was difficult since in this series a CT-based scala vestibuli position is missing. The coronal view shows diminishing of the fluid signal of the electrode (Fig. 4a,b) in comparison to the non-inserted cochlea (Fig. 3a,b). Coronal differentiation with a resolution of 0.8 mm between the scala tympani and scala vestibuli was difficult. A simultaneous DVT scan allowed for determination of the observed MRI-based position.

Discussion

The scalar position of the cochlear implant electrode is of high clinical importance, as it significantly influences the understanding of speech [1 9]. Therefore, post- or intraoperative estimation acts as a quality control for the surgery and is influenced by anatomy, electrode design and the surgeon's expertise.
The techniques used thus far for visual electrode assessment include the disadvantage of ionised radiation. The initial electrophysiological-based assessments of electrode position seem to be successful under acute and long-term conditions, but are electrode-dependent [2] and influenced by the brand-specific properties for intracochlear electrophysiological measurement.
MRI observations on cochlear implantees have been shown to be possible for all implant systems-with restrictions in terms of the field strength of the scanner and the need for a headband [10 11]. However, a persistent risk of magnet dislocation and a high rate of pain are known obstacles for some of the implant systems [3 4].
The introduction of a diametrically bipolar internal magnet solved the problem of magnet dislocation and pain [8]. Additionally, the observation of implant position-dependent artefact removal out of the cochlear and internal auditory canal area [5 6] allowed postoperative visual assessment of these otologically important regions.

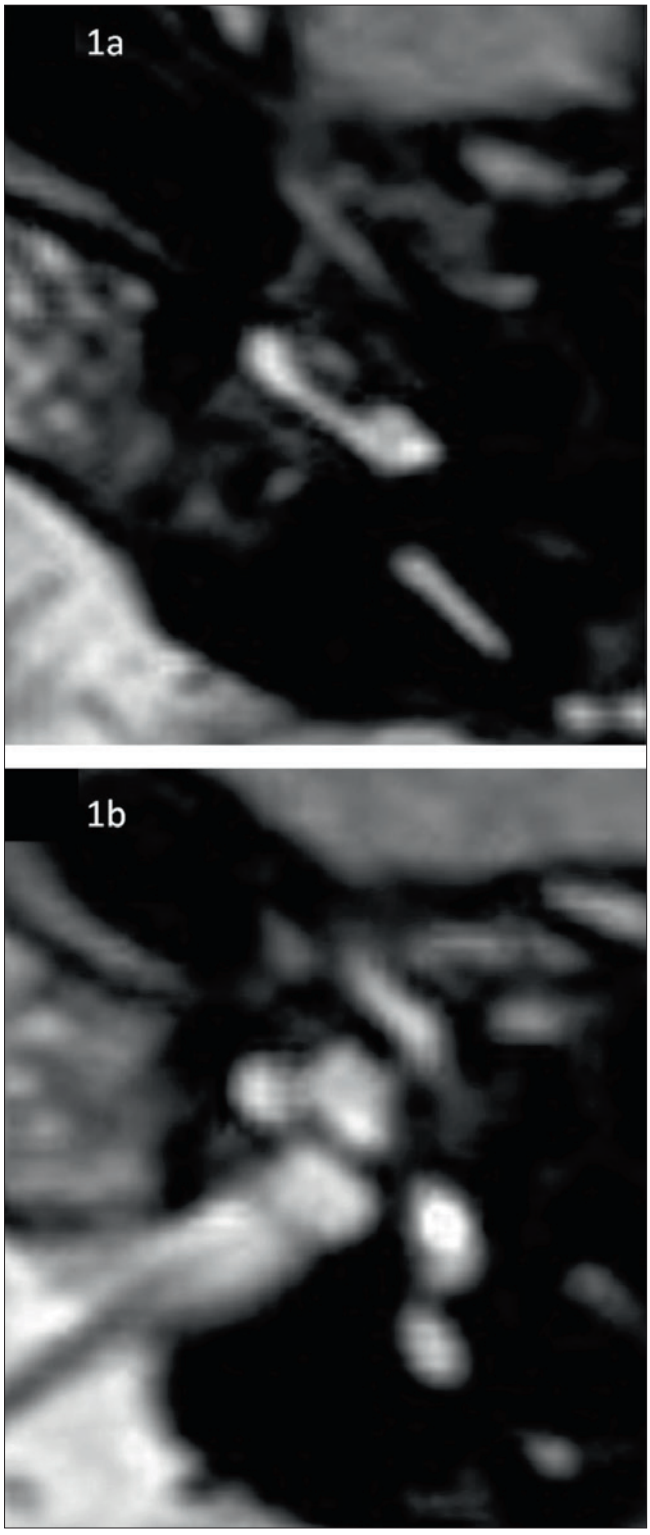

Fig. 1. 3T TSE t2 0.8 mm axial cochlea: a) basal turn, b) first turn.

In a previous study, it was shown that at 1.5T estimation of the electrode position by MRI is possible [7]. However, the electrodes used in this study were non-lateral wall

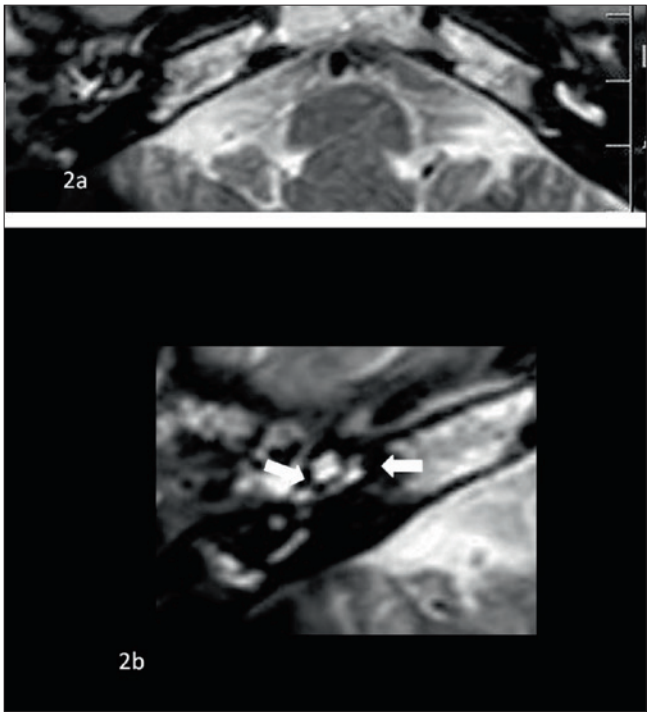

Fig. 2. 3T TSE t2 0.8 mm axial cochlea: a) basal turn inserted and not inserted, b) first turn with inserted electrode. Arrow indicates electrode-related signal diminishing.

electrodes. It is more difficult to visually assess the intracochlear position of lateral wall electrodes in DVT and FPT due to their high lateral position in the cochlear scala. In our study, we observed a positive correlation between CT-estimated positions and MRI observations. In all cases, we observed a scala tympani position in the basal turn. Compared to the previous 1.5 T study, the assessment in our study is characterised by higher visual resolution. This is related to the higher resolution of scans at 3T.

A difference between the fluid diminishing of the lateral wall electrode in this study and the perimodiolar/midmodiolar electrodes of the 1.5 T study was observed. While in both studies basal turn estimation of the electrode is easily possible, the more difficult assessment in the first turn is influenced by two factors: the scanner specific field strength and associated resolution abilities, and the electrode itself. Because the estimation of the electrode in the first turn is difficult in our 3T study and possible in the 1.5T study, electrode design thus seems to play a central role. Two points might explain the disadvantage of the lateral wall electrode used in terms of visual assessment. The first point is explained by a lower electrode volume of the lateral wall electrode in the first turn and therefore a lower fluid diminishing signal than a perimodiolar/midmodiolar electrode in this region.

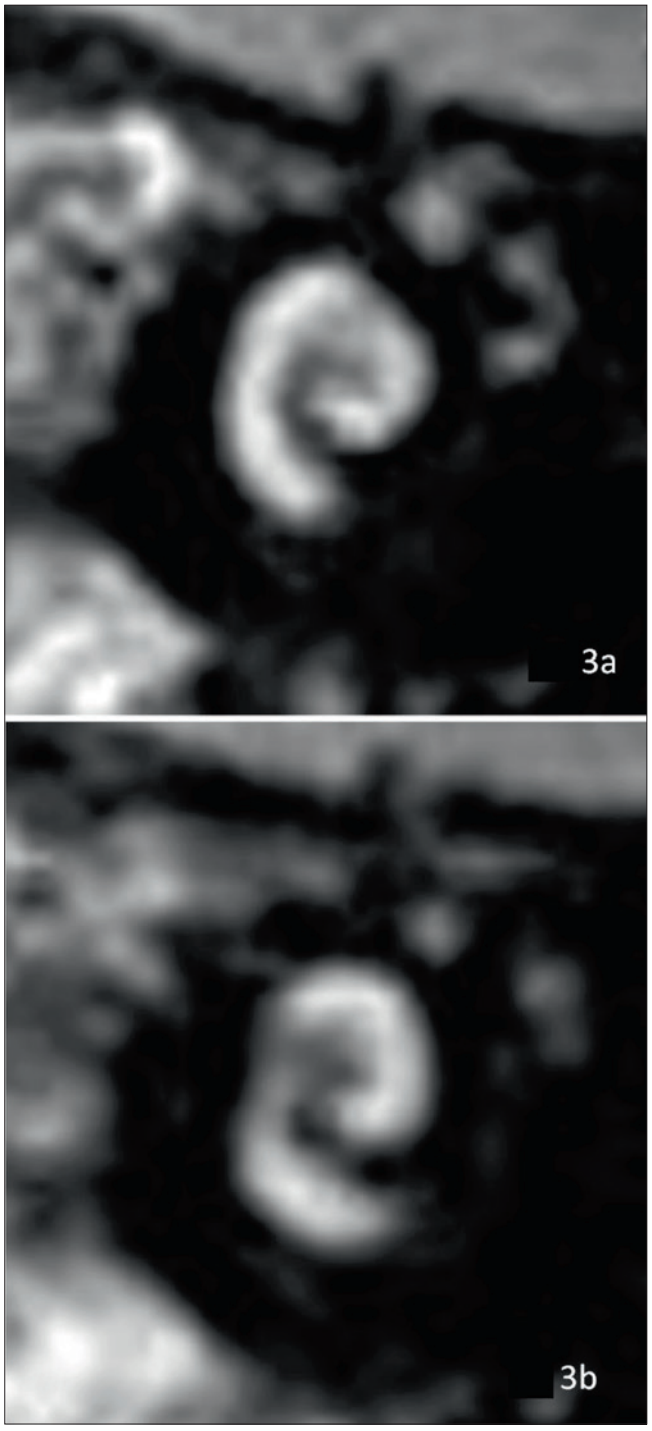

Fig. 3. 3T TSE t2 0.8 mm coronal cochlea: a) basal turn 1, b) basal turn 2.

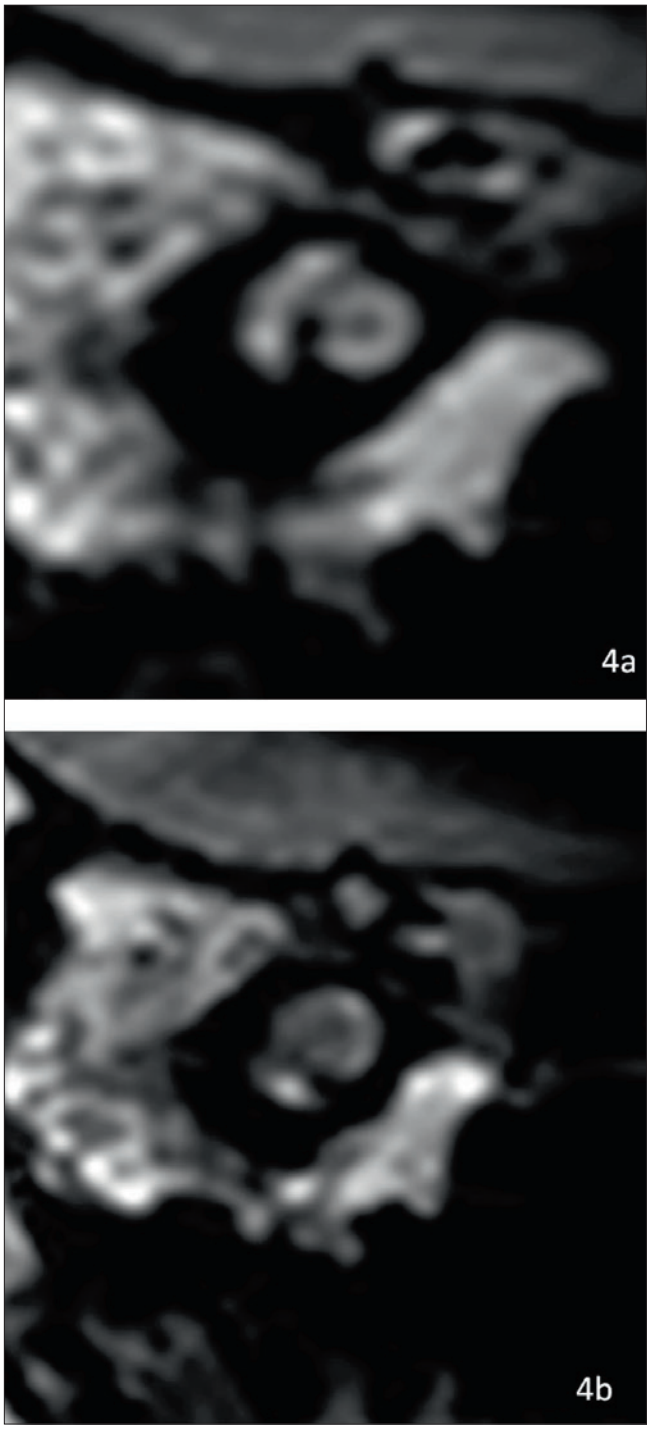

Fig. 4. 3T TSE t2 0.8 mm coronal cochlea: a) basal turn 1, b) basal turn 2 with inserted electrode.

The second point has to do with the lateral position of the electrode in the scala itself.

A limitation of the study is that a clear scala tympani to scala vestibuli translocation as a counterpart pattern for the scala tympani position was not observed. This is related to the lower translocation rate of lateral wall electrodes [12]. On the other hand, it is a disadvantage to the previously published 1.5T MRI study with a clear FTP and MRI-estimated visual translocation pattern of a perimodiolar electrode [7].

It can be assumed that with refined scanning protocols and prolonged scanning times better resolution will be possible.

Heating has a negative effect on neural structures when the temperature is increased to 43°C for more than 30 min [13]. The temperature increase of CI electrodes by a 3T scan is less than 3°C for 15 min scans. Usually the peak of temperature increase is reached during the first 3-5 min (personal communication, MEDEL, Innsbruck, Austria).

The postoperative MRI scanning at 3T allows high-resolution assessment of the internal auditory canal and the cochlea, if implanted with a diametrically bipolar internal magnet system. Ionised-free electrode assessment by MRI allows scanning in children. This opportunity is of reasonable clinical importance, even if it can be assumed that in a group of 400,000 CI-implanted patients, some developed a vestibular schwannoma after implantation. Another important reason for MRI scans in this patient group is the need for clarification of vertigo (e.g., infarction).

Therefore, the importance of postoperative MRI in the group of cochlear implantees with an otological and neurotological indication should not be underestimated.

Conclusions

Estimation of intracochlear position of a lateral wall cochlear implant electrodes by 3T MRI is possible for the basal turn. Electrode design plays a major role for visual assessment.

Acknowledgements

This study was supported by Medel, Innsbruck, Austria.

Conflict of interest statement

None declared.

References

1. Aschendorff A, Kubalek R, Turowski B, et al. *Quality control after cochlear implant surgery by means of rotational tomography*. Otol-Neurotol 2005;26:34-7.
2. Mittmann P, Todt I, Ernst A, et al. *Radiological and NRT-ratio-based estimation of slim straight cochlear implant electrode positions: a multicenter study*. Ann Otol Rhinol Laryngol 2017;126:73-8. https://doi.org/10.1177/0003489416675355.
3. Grupe G, Wagner J, Hofmann S, et al. *Prevalence and complications of MRI scans of cochlear implant patients*. HNO 2016;64:156-62. https://doi.org/10.1007/s00106-016-0128-8.
4. Hassepass F, Stabenau V, Maier W, et al. *Revision surgery due to magnet dislocation in cochlear implant patients: an emerging complication*. Otol Neurotol 2014;35:29-34. https://doi.org/10.1097/MAO.0b013e3182a5d2c5.
5. Walton J, Donnelly NP, Tam CY, et al. *MRI without magnet removal in neurofibromatosis type 2 patients with cochlear and auditory brainstem implants*. Otol Neurotol 2014;35:821-5. https://doi.org/10.1097/MAO.0000000000000330.
6. Todt I, Rademacher G, Mittmann P, et al. *MRI artifacts and cochlear implant positioning at 3 T in vivo*. Otol Neurotol 2015;36:972-6. https://doi.org/10.1097/MAO.0000000000000720.
7. Stratmann A, Mittmann P, Rademacher G, et al. *MRI-based estimation of scalar cochlear-implant electrode position*. Biomed Res Int 2017;2017, 6372704. https://doi.org/10.1155/2017/6372704.
8. Todt I, Tittel A, Ernst A, et al. *Pain Free 3 T MRI scans in cochlear implantees*. Otol Neurotol 2017;38:e401-4. https://doi.org/10.1097/MAO.0000000000001569.
9. Finley CC, Holden TA, Holden LK, et al. *Role of electrode placement as a contributor to variability in cochlear implant outcomes*. Otol Neurotol 2008;29:920-8. https://doi.org/10.1097/MAO.0b013e318184f492.
10. Crane BT, Gottschalk B, Kraut M, et al. *Magnetic resonance imaging at 1.5 T after cochlear implantation*. Otol Neurotol 2010;31:1215-20. https://doi.org/10.1097/MAO.0b013e3181ec1d61.
11. Kim BG, Kim JW, Park JJ, et al. *Adverse events and discomfort during magnetic resonance imaging in cochlear implant recipients*. JAMA Otolaryngol Head Neck Surg 2015;141:45-52. https://doi.org/10.1001/jamaoto.2014.2926.
12. Dhanasingh A, Jolly C. *An overview of cochlear implant electrode array designs*. Hear Res 2017;356:93-103. https://doi.org/10.1016/j.heares.2017.10.005.
13. Coffey RJ, Kalin R, Olsen JM. *Magnetic resonance imaging conditionally safe neurostimulation leads: investigation of the maximum safe lead tip temperature*. Neurosurgery 2014;74:215-24; discussion 224-5. https://doi.org/10.1227/NEU.0000000000000242.

Received: August 9, 2018 - Accepted: April 13, 2019

How to cite this article: Tek F, Müller S, Boga E, et al. *3T MRI-based estimation of scalar cochlear implant electrode position*. Acta Otorhinolaryngol Ital 2019;39:269-273. https://doi.org/10.14639/0392-100X-2309

Address for correspondence: Ingo Todt, Klinikum Bielefeld, Department of Otolaryngology, Head and Neck Surgery, Teutoburgerstr 50, 33604 Bielefeld, Germany. Tel. +49 521 581 3313. Fax +49 521 581 3303. E-mail: todt@gmx.net

In Memoriam of Antonio Pastore

On Thursday January 3, 2019 a serious and unexpected bereavement struck the University of Ferrara: Professor Antonio Pastore suddenly died.

Antonio was born in Nocera Inferiore on July 23, 1946. He moved with his family to Ferrara and after leaving the high school specialising in classical studies (maturità Liceo Classico), and graduated in Medicine and Surgery at the Estense University in 1973. His thesis was entitled "Electrocochleography".

This initial love for audiology will always follow him, even if his subsequent activity was mainly devoted to surgical oncology under the guidance of his Masters, the late lamented professors Gianpietro Teatini and Carlo Calearo.

His entire academic career took place at the Otorhinolaryngology Clinic of Ferrara, first as a Resident, then as an ordinary Assistant, Associate Professor and since 2003 as Full Professor. From 1998 to November 2016 he masterfully directed the clinic developing teaching, clinical and research activities.

He was a member of numerous Italian and International Scientific Societies and Associations and author of over 300 scientific papers. His main areas of interest were the study of neoplastic pathologies of the salivary glands in addition to audiology and laryngology.

I would like to remember Antonio Pastore, above all for the man he was.

A rigorous scientist in love with research and teaching, as witnessed by those who collaborated with him and the many fellow graduates under his leadership. He never forgot the respect for and comparison with students, and forever maintained a noble kindness in the relationship with patients to whom he always inspired confidence, courage and hope.

Outside the clinic he was a magnificent guest and an impeccable gentleman, always elegant and charismatic. For me he was a firm teacher but never authoritative. I owe a lot to him. He will remain in my memory and in the heart of all those who had the privilege of knowing him as an example and model of both professional and academic life.

Stefano Pelucchi

Notiziario SIO

CONSIGLIO DIRETTIVO SIO (2019-2020)

Bussi M.	Presidente SIO	(Scad. Maggio 2020)
Cuda D.	Vice-Presidente	(Scad. Maggio 2020)
Paludetti G.	Vice-Presidente	(Scad. Maggio 2021)
Succo G.	Segretario	(Scad. Maggio 2022)
Ruscito P.	Tesoriere	(Scad. Maggio 2022)
Vicini C.	Past President	(Scad. Maggio 2020)
Bruschini L.	Consigliere	(Scad. Maggio 2020)
Cocuzza S.	Consigliere	(Scad. Maggio 2020)
Da Mosto M.C.	Consigliere	(Scad. Maggio 2020)
Danesi G.	Consigliere	(Scad. Maggio 2020)
De Campora L.	Consigliere	(Scad. Maggio 2020)
Fiorella M.L.	Consigliere	(Scad. Maggio 2020)
Magnani M.	Consigliere	(Scad. Maggio 2020)
Miani C.	Consigliere	(Scad. Maggio 2020)
Panetti G.	Consigliere	(Scad. Maggio 2020)
Trimarchi M.	Consigliere	(Scad. Maggio 2020)
Della Vecchia L.	Coordinatore controllo qualità SIO	(Scad. Maggio 2021)
Capasso P.	Responsabile sito web SIO	(Scad. Maggio 2021)
Ralli G.	Bibliotecario	(Scad. Maggio 2021)
Ralli G.	Responsabile del Museo Storico	(Scad. Maggio 2021)
Di Santo D.	Segretario del Presidente	(Scad. Maggio 2020)
Pisani P.	Referente Privacy SIO	(Scad. Maggio 2021)

Collegio dei Probiviri SIO

Camaioni A.	(Scad. Maggio 2022)
Filipo R.	(Scad. Maggio 2022)
Leone C.A.	(Scad. Maggio 2022)
Serra A.	(Scad. Maggio 2022)
Spriano G.	(Scad. Maggio 2022)

Comitato Scientifico SIO

Coordinatore Comitato Scientifico SIO

Cassandro E. (Scad. Maggio 2021)

Segretario Comitato Scientifico SIO

Della Vecchia L. (Scad. Maggio 2021)

Membri Comitato Scientifico SIO

Bussi M.	Presidente SIO	(Scad. Maggio 2020)
Succo G.	Segretario SIO	(Scad. Maggio 2022)

Ansarin M.	Direttore responsabile Rivista	(Scad. Maggio 2022)
Cassandro E.	Comitato Audiol/Foniat/Vestibol	(Scad. Maggio 2021)
Camaioni A.	Comitato Laringologia	(Scad. Maggio 2021)
Castelnuovo P.	Comitato Rinologia	(Scad. Maggio 2021)
Passali D.	Comitato Dist. Resp. sonno	(Scad. Maggio 2021)
Piemonte M.	Comitato Day surgery	(Scad. Maggio 2021)
Danesi G.	Comitato Otologia/Otoneurologia	(Scad. Maggio 2021)
Nicolai P.	Comitato Oncologia/Ch. Ricostruttiva	(Scad. Maggio 2021)

Organo di Consulenza SIO

Ansarin M., Camaioni A., Casolino D., Cassandro E., Conticello S., Coppo L., De Benedetto M., De Campora E., Filipo R., Fiorella R., Laudadio P., Leone C.A., Nicolai P., Ottaviani A., Pagella F., Passali D., Paludetti G., Piemonte M., Puxeddu P., Radici M., Ralli G., Rinaldi Ceroni A., Sartoris A., Scasso F., Serra A., Sperati G., Spriano G.

Revisori dei conti

Salzano F.	(Scad. Maggio 2022)
Sessa M.	(Scad. Maggio 2022)

Comitato Scientifico ACTA

Alicandri Ciuffelli M. (AUORL)	(Scad. Maggio 2021)
Galli J. (AUORL)	(Scad. Maggio 2021)
Greco A. (AUORL)	(Scad. Maggio 2021)
Marioni G. (AUORL)	(Scad. Maggio 2021)
Piazza C. (AUORL)	(Scad. Maggio 2021)
Quaranta N. (AUORL)	(Scad. Maggio 2021)
Teggi R. (AUORL)	(Scad. Maggio 2021)
Testa D. (AUORL)	(Scad. Maggio 2021)
Bellocchi G. (AOOI)	(Scad. Maggio 2021)
Bertolin A. (AOOI)	(Scad. Maggio 2021)
Dispenza F. (AOOI)	(Scad. Maggio 2021)
Falcioni M. (AOOI)	(Scad. Maggio 2021)
Fiorino F. (AOOI)	(Scad. Maggio 2021)
Giourgos G. (AOOI)	(Scad. Maggio 2021)
Murri A. (AOOI)	(Scad. Maggio 2021)
Petrone P. (AOOI)	(Scad. Maggio 2021)

Comitato permanente per l'aggiornamento dello Statuto e del Regolamento

Bussi M.	Presidente SIO	(Scad. Maggio 2020)
Lauriello M.	Segretario dell'AUORL	(Scad. Maggio 2021)
Rugiu M.G	Segretario dell'AOOI	(Scad. Maggio 2020)
Succo G.	Segretario SIO	(Scad. Maggio 2022)
Bussi M., Gallo A., Quaranta N.	Colleghi nominati dall'AUORL	(Scad. Maggio 2021)
Ghidini A., Frosini P. Viti C.	Colleghi nominati dall'AOOI	(Scad. Maggio 2020)

Delegati

EUFOS	Marsella P.	(Scad. Maggio 2020)
	Marchioni D.	(Scad. Maggio 2021)
IFOS	Piemonte M.	(Scad. Maggio 2020)
	Galli J. (AUORL)	(Scad. Maggio 2021)
UEMS	Miani C.	(Scad. Maggio 2020)
	Motta Ga. (AUORL)	(Scad. Maggio 2021)
EHNS	Calabrese L.	(Scad. Maggio 2020)
	Motta Ga. (AUORL)	(Scad. Maggio 2021)

CONSIGLIO DIRETTIVO AOOI (2018-2020)

Radici M.	Presidente	(Scad. Maggio 2020)
Barbara M.	Presidente designato	(2020-2022)
Bellocchi G.	Vice-Presidente	(Scad. Maggio 2020)
Della Vecchia L.	Vice-Presidente	(Scad. Maggio 2020)
Tortoriello G.	Vice-Presidente	(Scad. Maggio 2020)
Rugiu M.G.	Segretario-tesoriere	(Scad. Maggio 2020)
Bertolin A.	Consigliere	(Scad. Maggio 2020)
Crosetti E.	Consigliere	(Scad. Maggio 2020)
Capasso P.	Consigliere	(Scad. Maggio 2020)
Caporale C.	Consigliere	(Scad. Maggio 2020)
De Vito A.	Consigliere	(Scad. Maggio 2020)
Franzetti A.	Consigliere	(Scad. Maggio 2020)
Galfano M.	Consigliere	(Scad. Maggio 2020)
Nardone M.	Consigliere	(Scad. Maggio 2020)
Palladino R.	Consigliere	(Scad. Maggio 2020)

Collegio dei probiviri

Barbieri F.	(Scad. Maggio 2020)
De Benedetto M.	(Scad. Maggio 2020)
Laudadio P.	(Scad. Maggio 2020)
Richichi M.	(Scad. Maggio 2020)
Villari G.	(Scad. Maggio 2020)

CONSIGLIO DIRETTIVO AUORL (2019-2021)

Nicolai P.	Presidente
de Vincentiis M.	Vice-Presidente
Lauriello M.	Segretario-tesoriere
Benazzo M.	Consigliere
Berrettini S.	Consigliere
Iengo M.	Consigliere
Presutti L.	Consigliere
Canale A.	Consigliere
La Mantia I.	Consigliere
Rigante M.	Consigliere
Carta F.	Consigliere
Paludetti G.	Past-President

Revisori dei conti (2017-2021)

Cocuzza S.
Pelucchi S.

Collegio dei probiviri (2019-2021)

Conticello S.
Gaini R.
Martini A.
Passali D.
Pia F.

Comitato permanente per l'aggiornamento dello Statuto e del Regolamento (2017-2021)

Bussi M.
Iengo M.
Ricci G.

Temi scientifici 2020-2021

Congresso Nazionale SIO 2020
Relazione ufficiale
"La malattia metastatica in oncologia testa e collo" *a cura di Paolo Pisani*
Tavola Rotonda ufficiale
"Le paralisi periferiche del nervo faciale: diagnosi, terapia e riabilitazione" *a cura di Livio Presutti*

Relazione ufficiale SIO 2021
"Patologie di confine in Otorinolaringoiatria e loro trattamento" *a cura di Gaetano Paludetti*