Salivary flow decreases in healthy elderly people independently of dental status and drug intake

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Résumé:

In humans, oral food consumption is by far the most important point where food's organoleptic properties can be perceived and can elicit sensory pleasure. It is also the ultimate stage of the food supply chain and the beginning of the food disintegration and digestion process. However, in regard to the influence of ageing on food oral processing, this topic has been mainly investigating through mastication, whereas salivation remains largely unexplored. The present experiment aimed at studying the impact of normal ageing on salivary flow taking into account the dental status and the number of drugs taken by the elderly people. This was achieved by comparing resting and stimulated salivary flows of young versus healthy elderly adults (*i.e.*, autonomous elderly people without acute pathology). Ninety-three young adults (22-55 years old) and 84 elderly people (70-92 years old) underwent a measurement of resting and stimulated salivary flows and an oral examination (teeth counting; functional unit counting i.e., counting occluding tooth pairs). The present study showed an average 38.5% reduction of resting salivary flow and 38.0% reduction of stimulated salivary flow in healthy elderly people compared to young adults. This reduction was observed independently of the dental status and drug intake: elderly people presented reduced salivary flow even if they did not take any drugs or if their dental status was similar to the one of the young adults. The results also highlight a large inter-individual variability both in young and elderly adults.

Practical applications

During oral food consumption, saliva plays a key role in the acceptance of food and beverage by modulating the perception of texture, taste and aroma, as well as providing eating comfort by assisting the food breakdown process into a bolus that can be safely swallowed. However, in regard to the influence of ageing on food oral processing, the present results demonstrate a reduced salivary flow in healthy elderly people. Consequently, there is a need for developing foods tailored to the salivary capacities of elderly people aside from the efforts put into the development of foods tailored to the mastication and swallowing abilities of this population. In fact, in the context of an ageing population, the development of products meeting an elderly person's functional capacities becomes a major challenge for the food industry as well as for society.

INTRODUCTION

In humans, oral food consumption is by far the most important point where food's organoleptic properties (texture, taste, and aroma) can be perceived and elicit sensory pleasure. It is also the ultimate stage of the food supply chain and the beginning of the food disintegration and digestion process (Chen, 2009). The different mechanisms involved in oral food consumption, which is referred to as "food oral processing", depend both on the food's structure and the individual's oral physiology (Salles *et al.*, 2011). However, in regard to the influence of ageing on food oral processing, this topic has been mainly investigating through mastication whereas salivation remains largely unexplored.

Saliva is a physiological fluid that plays a crucial role in preserving and maintaining oral health and eating comfort (Carpenter, 2012). Saliva is secreted by three major salivary glands (submandibular, sublingual and parotid glands) and several minor salivary glands located all over the oral cavity (Christensen, 1986). During oral food consumption, saliva has three major functions. First, saliva plays a key role in the acceptance of food and beverage by modulating the perception of oral sensations (taste, viscosity, smoothness, juiciness, astringency, etc.) and aroma release. In fact, taste compounds should be in an aqueous solution to reach and activate taste buds (Fischer *et al.*, 1994), and it has been demonstrated that taste sensitivity is related to saliva composition (Dsamou *et al.*, 2012). Furthermore, texture perception is influenced by saliva composition. Engelen *et al.* (2007) have demonstrated that subjects with a high α-amylase activity had a decreased thickness perception of a starch-based custard. Finally, saliva can impact aroma release by assisting in the food breakdown process, by retaining or releasing aroma compounds depending on their affinity with saliva, and by inducing chemical reactions likely to produce new volatile compounds (Gierczynski *et al.*, 2011). Second, saliva, as well as mastication, transforms a food sample into a bolus that can be safely swallowed (Prinz and Lucas, 1997). The water in saliva moistens the food particles, whereas the salivary mucins bind masticated food into a coherent and slippery bolus that can easily slide through the oesophagus without damaging the mucosa. Saliva enzymes also initiate the digestion of carbohydrates and triglycerides in the food bolus (Salt and Schenker, 1976; Hamosh and Burns, 1977). Third, saliva dilutes and removes substances from the oral cavity after swallowing ("oral clearance"; Lagerlof and Oliveby, 1994; Lenander-Lumikari and Loimaranta, 2000). In fact, salivation and swallowing are acknowledged to be important processes for eliminating injurious and noxious agents and bacteria from the oral cavity (Pedersen *et al.*, 2002). Saliva clears sugar and acids from the oral cavity and thereby protects teeth from erosion. Finally, teeth and mucous membranes are covered by a protective film of saliva, which prevents the occurrence of caries (Ericsson, 1953).

In elderly people, it has been demonstrated that the cumulative effect of ageing and associated changes, such as tooth loss (Dormenval *et al.*, 1998; Srinivasulu *et al.*, 2014), drug intake (Handelman *et al.*, 1989; Bardow *et al.*, 2001; Johanson *et al.*, 2015; Thomson, 2015) and disease (Ship *et al.*, 1990; Ship, 1992; Ship and Puckett, 1994; Chu *et al.*, 2015), may affect salivary flow. However, to the best of our knowledge, very few studies ~~(if not)~~have investigated the impact of normal ageing on salivary flow taking into account the dental status and the number of drugs taken by the elderly people. In these studies, many methods were used to measure the salivary flow. Therefore, a comparison of the results is difficult to make. Moreover, none of the studies have explored the relationship between dental status and salivary flow in elderly people.

Consequently, the present experiment aimed at studying the impact of normal ageing on salivary flow taking into account dental status and the number of drugs taken by the elderly

people. This was achieved by comparing resting and stimulated salivary flows of young versus healthy elderly adults (*i.e.*, autonomous elderly people without acute pathology).

MATERIALS AND METHODS

Participants

Young adult panel. A panel of young adult volunteers (n=93) was recruited in Dijon during a period of two months. The recruitment criteria were the following: aged between 20 and 55 years and good dental status (no missing teeth except the third molar, no occlusion disorders, and no daily drug intake). An interview was carried out with each volunteer to ensure that they met the inclusion criteria.

Elderly adult panel. The data were collected as part of a programme aimed at studying the relationship between oral health and eating behaviour (AlimaSSenS project: towards an adapted and healthy food supply for elderly people). A panel of elderly volunteers (n=84) was recruited from a population of elderly people living at home in Dijon during a period of six months. The recruitment criteria were the following: older than 70 years old, no acute pathological episodes at the time of the experiment, and scoring at least 24 on the Mini Mental State Evaluation (MMSE) (Folstein *et al.* 1975). An interview was carried out with each volunteer to ensure that they met the inclusion criteria.

Procedure

Young adult volunteers underwent a salivary flow test (resting and stimulated). The session was organized as a face-to-face interview that was conducted by 3 experimenters who had previously participated in a 1-day training session.

Elderly adult volunteers took part in one session with a duration of approximately one hour and thirty minutes. During this session, participants completed the Geriatric Oral Health Assessment Index (GOHAI), which is a questionnaire that evaluates self-perception of oral health (Slade and Spencer, 1994; Tubert-Jeannin *et al.*, 2003). The participants were also interviewed on their food habits and drug intake. Then, the participants sat in an articulated resting chair, and a trained experimenter carried out the oral examination (teeth counting and functional unit counting). Finally, the participants completed the xerostomia questionnaire (Thomson *et al.*, 1999) and performed a measure of salivary flow (resting and stimulated). The sessions were organized as face-to-face interviews that were conducted by 3 experimenters ~~(all women)~~ who had previously participated in a 4-hour long training session.

Measurements

Salivary flow. Resting and stimulated salivary flows were measured as previously described (Feron *et al.*, 2014; Neyraud *et al.*, 2012). The participants were asked not to smoke, eat or drink at least one hour before collecting the saliva. Resting salivary flow was measured by instructing the participant to spit out the saliva into a pre-weighed screw-cap cup every time they felt like swallowing over a period of 5 minutes for the young adults and 10 minutes for the elderly participants. Stimulated salivary flow was measured by instructing the participants to masticate a piece of pre-weighed parafilm while spitting out the saliva into a pre-weighed screw-cap cup every time they felt like swallowing over a period of 5 minutes. Cups were weighed, and salivary flow rates were expressed in ml/min assuming that one g of saliva corresponds to one ml.

Teeth counting. A trained experimenter counted the number of natural, restored and fixed prosthetic teeth (participants who wore dentures were asked to remove them for this measure).

Functional unit counting. A functional unit was defined as a pair of posterior antagonist teeth that had at least one contact area during chewing. The number of functional units was evaluated by asking the participants to chew 1-2 cycles on 200-µm thick articulating paper; the number of teeth on the mandibular arch that had at least one colour mark provided the number of functional units. The participants with dentures were asked if they had used their dentures while eating during their three last meals. Those who had not done so were asked to remove their dentures before completing this measure.

Data analysis

Student's *t*-tests were performed using the TTEST procedure provided in the SAS software (SAS Institute INC., Cary, NC, USA). Equality of variance was first assessed by using a folded form of the F statistic (Steel and Robert, 1980). Depending on the results, groups were compared using the *t* statistic when the variances were equal or the Cochran and Cox (1950) approximation when the variances were unequal. Analyses of variance (ANOVA) were done using the GLM procedure provided in the SAS software (ss3 option). Post-hoc analyses were performed using the Student Newman Keuls test. Means (M) are associated with the standard error of the mean (SEM). The threshold for significance was set to 5%.

RESULTS

Ninety-three young adults (48 women and 45 men) and 84 elderly participants were included (47 men and 37 women). No significant differences were observed regarding sex distribution (χ^2=1.01; ns). Characteristics of the participants are summarized in the **Table 1**. As expected, the dental status of the young participants was better than that of the elderly participants (number of missing teeth for young adults: 0).

Table 1 about here

Impact of ageing on salivary flow

Figure 1 presents the box-plot distributions of the resting and stimulated salivary flow for the young and elderly panels. The box-plot distributions reveal a median difference but also a larger variability for the young adult participants for both the resting and stimulated salivary flow. Actually, variances in age groups were significantly unequal for the stimulated flow [$F(92,83)=2.11$; $p<0.001$] but not for the resting flow [$F(92,83)=1.35$; $p=0.17$]. Both the resting and stimulated salivary flow were lower in the elderly participants than in the young participants [resting: $t(175)=6.00$; $p<0.001$; stimulated: $t(164)=6.91$; $p<0.001$].

With regard to resting salivary flow, no impact of sex was observed for the elderly participants [$t(78)=-0.30$; $p=0.76$; M= 0.30 ml/min ± 0.03; M= 0.31 ml/min ± 0.03, for elderly women and elderly men, respectively], while a tendency was observed for the young participants [$t(90)=-1.76$; $p=0.08$], with young women tending to have a lower resting flow [M=0.46 ml/min ± 0.03] than young men [M=0.54 ml/min ± 0.03]. With regard to stimulated salivary flow, a significant impact of sex was observed for both the young [$t(86)=-2.15$; $p<0.05$] and elderly participants [$t(82)=-2.20$; $p<0.05$], with women having a reduced stimulated flow [young: M=2.24 ml/min ± 0.14; elderly: M=1.34 ml/min ± 0.10] compared with men [young: M=2.70 ml/min ± 0.17; elderly: M=1.67 ml/min ± 0.12].

Figure 1 about here

Impact of ageing versus dental status and drug intake on salivary flow

To further study the impact of dental status, two sub-groups were considered among the elderly participants: elderly participants with a good dental status, which had 7 or more posterior functional units and no dentures (n=27), and elderly participants with a poor dental status, which had 4 or fewer posterior functional units (n=19). The thresholds of 7 and 4 functional units to define good and bad dental status, respectively, were defined according to Leake *et al.* (1994). It should be noted that all 93 young adults presented 7 or more posterior functional units. **Figure 2** presents the resting and stimulated salivary flow for the young adults and each elderly dental group. The ANOVA revealed a significant group effect (resting: $F(2,136)=16.40$; $p<0.001$; stimulated: $F(2,136)=16.11$; $p<0.001$). According to the post-hoc analyses, the salivary flows of young adults were significantly greater than the salivary flows of elderly people regardless of their dental status. No significant effect of dental status was observed in the elderly participants.

Figure 2

To investigate the impact of drug intake, two subgroups were considered among the elderly participants: elderly participants not taking any drugs (n=19) and elderly participants taking at least 4 or more drugs per day (n=28). The thresholds of intake of 4 or more drugs per day was defined according to Handelman *et al.* (1989) who showed a significant decrease in stimulated salivary in elderly people taking four drugs per day. It should be noted that none of the 93 young adults took drugs during the time of the experiment. **Figure 3** presents resting and stimulated salivary flow for young adults and each elderly drug intake group. The ANOVA revealed a significant group effect (resting: $F(2,137)=13.02$; $p<0.001$; stimulated: $F(2,137)=16.07$; $p<0.001$). According to the post-hoc analyses, the salivary flows of young adults were significantly greater than salivary flows of elderly people whether they took drugs or not. No significant effect of drug intake was observed within the elderly population recruited for this study.

Figure 3

DISCUSSION

The present study showed, on average, a 38.5% reduction in the resting salivary flow and 38.0% for the stimulated salivary flow in healthy elderly people compared with young adults. This reduction was observed independently of drug intake and the dental status: elderly people presented a reduced salivary flow even if they did not take any drugs or if their dental status was similar to the one of the young adults. These results support the results of Affoo *et al.* (2015) who recently conducted a meta-analysis on the impact of ageing on salivary flow. According to Affoo *et al.*, ageing is associated with reduced salivary flow that cannot be fully explained by drug intake or disease (dental status was not explored). In fact, from a biological standpoint, ageing is accompanied by structural changes in the salivary glands, such as a reduction in acinar volume, a loss of secretory tissue and an increase of adiposity (Scott, 1986; Scott *et al.*, 1987). Furthermore, it has been suggested that age-related neurophysiological changes may account for the changes in salivary secretion (Baum, 1987). However, further studies are needed to clarify the link between these changes and saliva secretion in elderly people and to uncover the mechanisms beyond the impact of ageing *per se* on salivary flow.

Beyond these age-related processes, it has been demonstrated that drug intake may have a strong impact on saliva flow (Handelman *et al.*, 1989; Bardow *et al.*, 2001; Johanson *et al.*, 2015; Thomson, 2015). According to Sreebny and Schwartz (1997), 42 drug categories and 56 subcategories are known to be xerogenic, *i.e.*, inhibit saliva secretion through various pathways. In fact, Handelman *et al.* (1989) observed that participants who took more than three drugs per day had a stimulated salivary flow significantly less than participants who did not take any drugs. However, in the present experiment, no difference was observed between elderly people who did not take any drugs and elderly people who took more than three drugs per day. A similar result was previously observed by Nagler and Hershkovich (2005) who observed no significant difference in the resting salivary flow between drug-free elderly participants and elderly people who were taking drugs. This study included 28 subjects aged between 60 and 90 years old with an average age of 75.8 years with 14 participants that were drug free and 14 participants that consumed drugs (patients treated routinely).

In the same way, previous studies have demonstrated a correlation between the number of remaining teeth and a reduction in the salivary flow in an elderly population (Ikebe *et al.*, 2011; Samnieng *et al.*, 2012). Interestingly, whereas some authors have hypothesized that the age-related decline in bite strength may result in less stimulation of salivary glands and, thus, in a reduced salivary flow (Affoo *et al.*, 2015). Other authors have demonstrated that elderly people with a low salivary flow rate were more likely to lose teeth (Caplan and Hunt, 1996). However, in the present experiment, no correlation was observed between the dental status and the saliva flow. Furthermore, Flink *et al.* (2008) showed a relation only in the case of severely reduced salivary flow among women over 50 years old. The author proposed that this link was mainly explained by the development of caries due to a lack of saliva secretion and, thus, protection.

The present study included "healthy" elderly people: older adults who lived independently at home, who did not suffer from an acute pathology (e.g., no cancer) or dementia (e.g., Alzheimer' disease) and who had a good nutritional status. Indeed, the mean MMSE score for the present sample was 28.7 out of 30; no participant scored below 25 (the MMSE screens for cognitive impairment; scores that are greater than or equal to 25 points indicate normal cognition). The mean MNA score of the present sample was 27.3 out of 30; only two participants were at risk for malnutrition (the MNA screens for malnutrition; scores that are greater than or equal to 24 points indicate normal nutritional status; scores that range from 17 to 23.5 indicate a risk of malnutrition, and scores below 17 points indicate malnutrition). This means that these participants were free from any pathology. In fact, 65 of the participants took an average of 3.6 drugs per day mainly for chronic diseases, such as hypertension. However, the present sample may be quite free of confounding factors that are often associated with drug intake and/or poor dental status, such as poor nutritional status, decline in general health, onset of neurological disorders. This question of confounding factors was evidenced in the study conducted by Flink *et al.* (2008) on 1247 volunteers and considered similar variables as the ones in the present study. The authors showed that, for instance, in young adults (less than 50 years old) drug intake did not explain the decrease in salivary flow. The authors suggest the involvement of other variables, such as body mass index greater than 25 kg/m^2, malnutrition and disease. For instance, the authors showed that the only variable that explained a very low stimulated salivary flow in a subset of this population was a diagnosed disease. Conversely, in the same study, the effect of drugs on salivary flow was demonstrated in the older population (65 to 69 years old), but in this population, an average of 52% of women and 58% of men with a disease had poor general health.

Finally, the present experiment highlighted the large inter-individual variability both in the young and elderly panel. Among the elderly panel, 13% of the participants were suffering from hyposalivation (resting salivary flow less than 0.1 ml/min), whereas 18% of the participants had a salivary flow greater than the mean salivary flow of the young panel. A similar conclusion was performed by Flink *et al.* (2008). In their best model predicting a very low unstimulated salivary flow rate, the independent variables explained only 10% of the difference between individuals. Two types of factors can be proposed to account for the variability observed in the elderly panel for the salivary flow **(Figure 4)**:

- Life-style factors, such as diet or smoking habits. In fact, Ernest (1993) showed a positive relationship between flow rate and the intake of 18 out of 22 nutrients, with a highly significant correlation for calories, protein, carbohydrates and vitamin C and B-6, amongst others. Moreover, it has been demonstrated that a modification in diet (liquid, less acidogenic or firmer texture) can either increase or decrease the salivary flow (Dodds *et al.*, 2005). Regarding smoking habits, the salivary flow rate was significantly reduced in long-term smokers (mean salivary flow: 0.38 ml/min) compared to non-smokers (0.56 ml/min) (Rad *et al.*, 2010).
- Ageing factors including ageing-related processes, such as the hydration status (Buffa et al., 2011), structural changes in the salivary glands, neurophysiological changes or ageing-related events, such as the onset of a pathology or dementia, may impact the salivary flow directly or indirectly through drug intake.

However, longitudinal studies are expected to decipher the correlation between the salivary flow of young and older adults, and, namely, whether individuals who have the greatest salivary flows when they are young will have reduced but still the greatest salivary flows when older compared with their age group. Furthermore, studies are expected to disentangle the relative impact of each factor on the reduction of the salivary flow in an elderly population.

Ethical Statements

The authors declare that they do not have any conflicts of interest. For the young panel, the experimental protocol was approved by the French Ethics Committee for Research (Comité de Protection des Personnes Est-1 N°2008/15) and by the Direction Générale de la Santé, France (N° DGS2008-0196). For the elderly panel, the experimental protocol was approved by the French Ethics Committee for Research (CPP Est III, Nancy, #15.04.04, ANSM #2015-A00279-40). In accordance with ethical standards, all participants received written and oral information on the study before signing a consent form.

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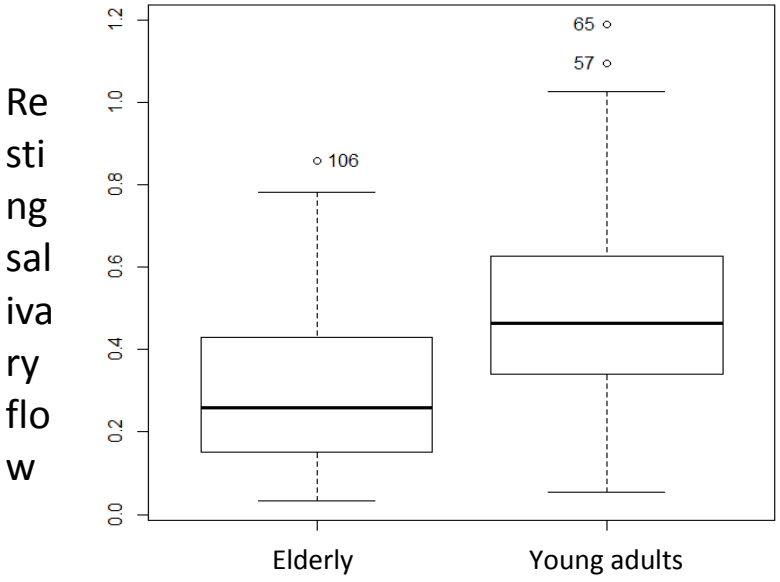
Table 1. Characteristics of the young and elderly panels.

	Young panel (n=93)			Elderly panel (n=84)		
	M	SEM	Range	M	SEM	Range
Age	38.94	8.37	22 - 55	76.19	4.63	70 - 92
Number of teeth				21.40	9.00	0 - 32
Number of functional units				6.02	2.06	0 - 10
Resting salivary flow	0.50	0.23	0.05 – 1.19	0.31	0.19	0.03 - 0.86
Stimulated salivary flow	2.47	1.06	0.70 – 5.45	1.52	0.73	0.11 - 4.01

M: mean; SEM: standard error of the mean.

FIGURE 1

Box-plot distributions of the resting (1.a) and stimulated (1.b) salivary flow for the young and elderly panels. The bottom and the top of the box correspond to the 25th and 75th percentile, respectively. The thick band corresponds to the median. The ends of the whiskers represent the lowest/highest data still within the 1.5 interquartile range. Any data points not included between the whiskers are plotted as outliers with a dot.



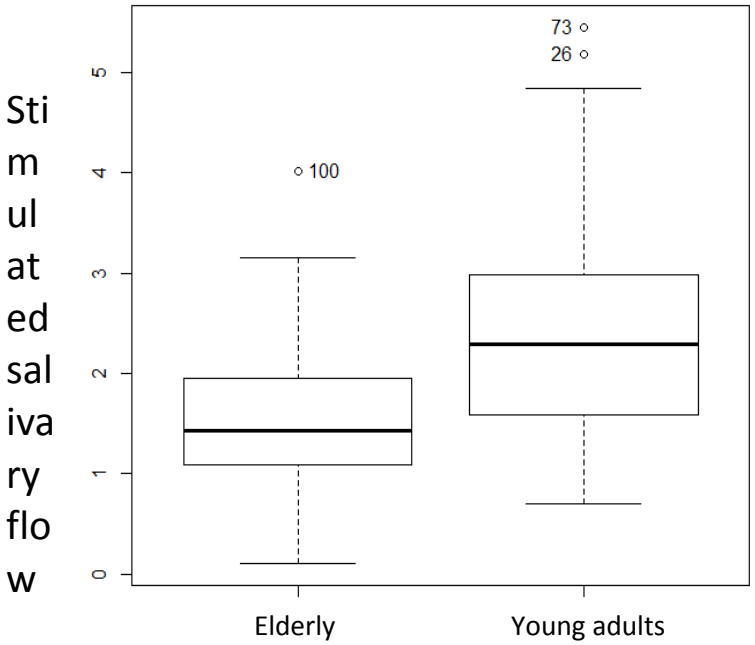
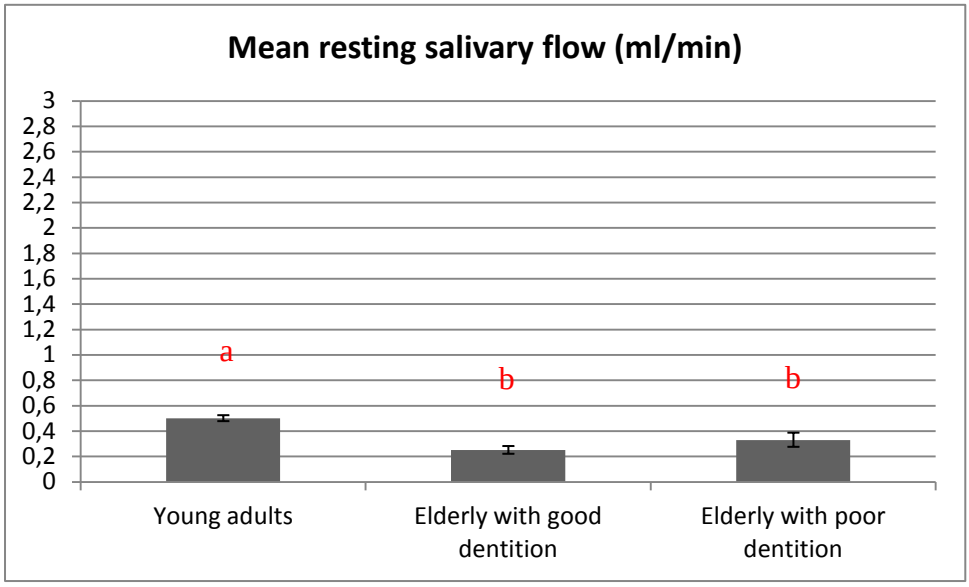


FIGURE 2

Representation of the resting (2.a) and stimulated (2.b) salivary flows for young adults, elderly with good dentition and elderly with poor dentition. The error bars correspond to the standard error of the mean. For each variable, the means with the same letter are not significantly different ($p<.05$). Resting (2.a): M_{young}= 0.50 ml/min ± 0.02; $M_{elderly\ good\ dentition}$= 0.24 ml/min ± 0.03; $M_{elderly\ poor\ dentition}$= 0.33 ml/min ± 0.06. Stimulated (2.b): M_{young}= 02.46 ml/min ± 0.11; $M_{elderly\ good\ dentition}$= 1.53 ml/min ± 0.11; $M_{elderly\ poor\ dentition}$= 1.42 ml/min ± 0.19.

2.a



2.b

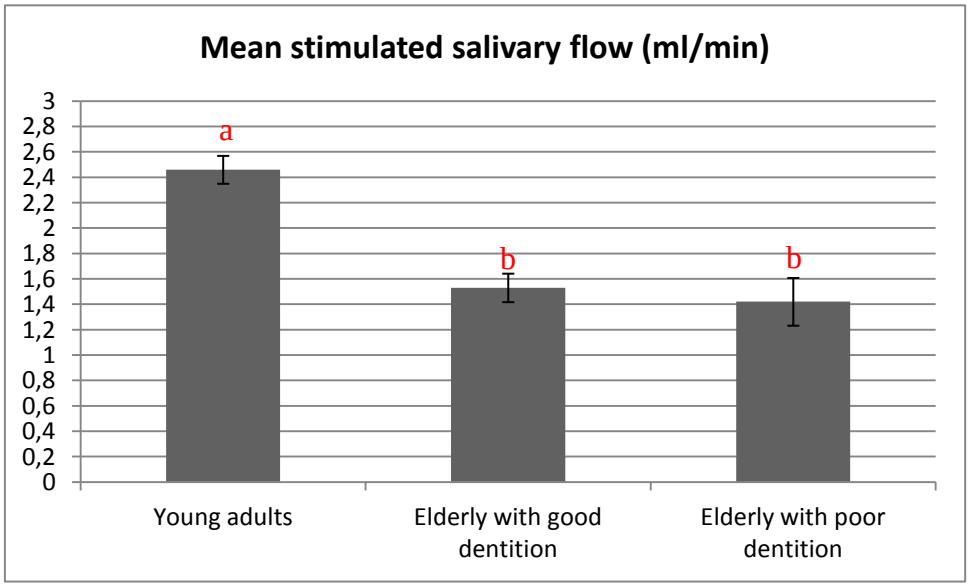
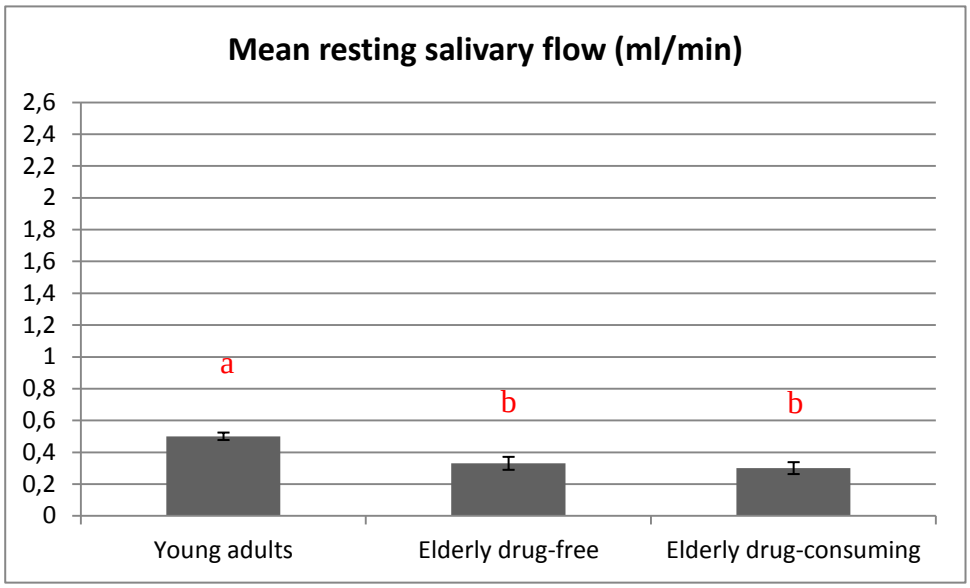


FIGURE 3

Representation of the resting (3.a) and stimulated (3.b) salivary flows for young adults, elderly drug free and elderly consuming drugs. The error bars correspond to the standard error of the mean. For each variable, the means with the same letter are not significantly different ($p<.05$). Resting (3.a): M_{young}= 0.50 ml/min ± 0.02; $M_{elderly\ drug\ free}$= 0.33 ml/min ± 0.04; $M_{elderly\ drug\ intake}$= 0.30 ml/min ± 0.02. Stimulated (3.b): M_{young}= 02.46 ml/min ± 0.11; $M_{elderly\ drug\ free}$= 1.45 ml/min ± 0.13; $M_{elderly\ drug\ intake}$= 1.55 ml/min ± 0.10.

3.a



3.b

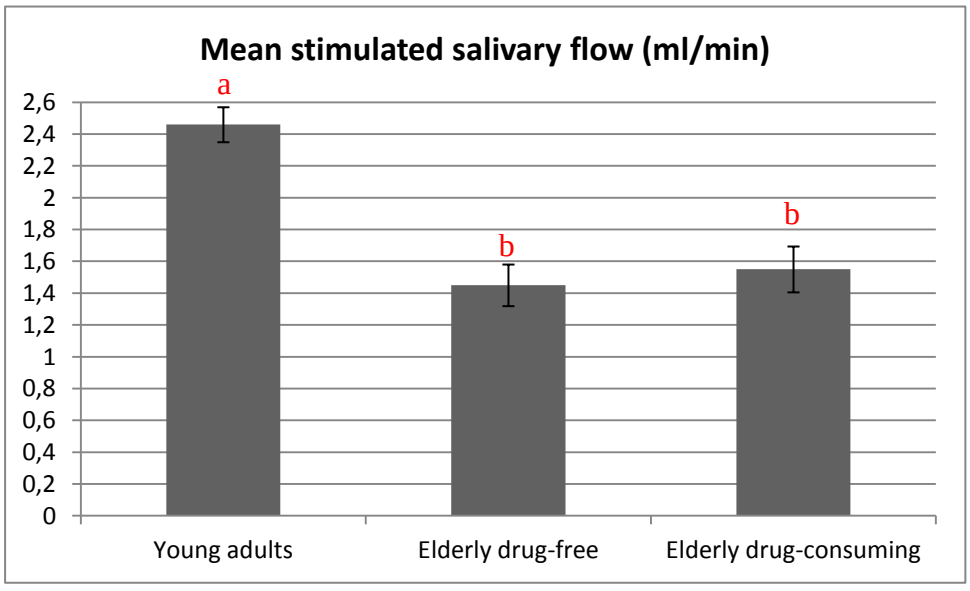


FIGURE 4

Representation of the factors likely to explain the variability observed in the elderly panel with respect to the salivary flow.