Perfumery and the Sixth Sense

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"Humans have a complete set of organs which are traditionally described as non-functional, but which, if seen in any other mammal, would be recognized as part of a pheromone system."

A. Comfort[1]

Introduction

The role of the vomeronasal organ (VNO) as the pheromone detection system in rats was first demonstrated by Johns, et al.[2] These findings have been confirmed, and generalized to other mammals.[3]

The human VNO was discovered by Ruysch in 1703.[4] Since that time there have been many reports; some confirm Ruysch, while others state that in adult humans the VNO is nonexistent, vestigial or atrophic. It is only recently that several different research groups, using more extensive investigations, have established unequivocally that the VNO is present in all normal humans from neonates to adults.[5-7] In addition, histochemical techniques using specific neuronal markers have demonstrated the presence of neuroepithelial cells in the human VNO.[8] The bilateral location of the vomeronasal organ in the human nose (Figure 1) ensures access to substances present in inspired air.

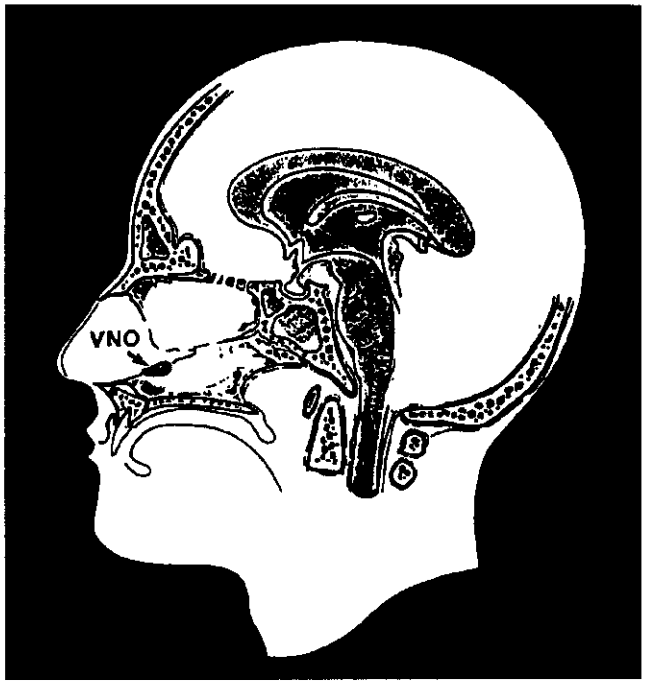

Figure 1. Location of the human vomeronasal organ

The functional nature of the human VNO was described in 1991.[9] Substances present in human skin were found to activate the human VNO in a species-specific and sexually dimorphic fashion. Thus our "sixth sense" is actually a real sensory system providing us with another window on the world.

The definition of pheromones includes that they are external chemical messengers secreted to the outside by an individual and received by a second individual of the same species.[10] Thus human pheromones, by definition, will be found secreted to the outside of a human individual. This paper describes some investigations of the chemistry and physiology of this class of substance, and the significance to perfumery.

Methods

The compounds tested are, for the most part, commercially available and were used as such after checking purity by thin layer chromatography. The syntheses of some less commonly available test substances have been described elsewhere.[11]

The most direct noninvasive method for measuring the activity of substances in the human VNO is to record the electrogram from this organ (electrovomeronasogram or EVG). This is accomplished by placing a "multifunctional miniprobe" in the human VNO. The miniprobe delivers a discrete pulse of test substance, dispersed in an air stream, to the VNO. The air stream is confined by means of vacuum aspiration to impinge on a small surface (diameter ≅ 1 mm),

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and the summated local potential is detected by means of an electrode. The amplitude of the resulting EVG is measured in millivolts. The experimental methodology has been described elsewhere.[9,12]

Results: Skin Steroids

A selection of various human skin substances and analogs was presented to the human male VNO, and EVGs were recorded. The results are shown in Figure 2. The same selection was also presented to the human female VNO, and the results are shown in Figure 3.

The most obvious feature of these results is that only two of these compounds are significant stimulators of the human VNO. The substances are estra-1,3,5(10),16-tetraen-3-ol (estratetraenol) and androsta-4,16-dien-3-one (androstadienone) (Figure 4). Androstadienone is a strong odorant.[13] However, the scavenging system of the miniprobe prevents contamination of the olfactory epithelium, so no odor is detected during these experiments.

A noteworthy feature of estratetraenol is that it is completely devoid of odor.[14,15] This belies the idea that pheromones should be odorous, a misconception repeated in the pages of this journal as recently as 1993.[16] Such an idea only makes sense if the existence of VNO is ignored.

Another obvious feature of the data shown in Figures 2 and 3 is that the VNO responses to these substances show a high degree of gender specificity. Here, it is noteworthy that men are particularly sensitive to estratetraenol, and women are particularly sensitive to androstadienone.

A third feature of the data shown in Figures 2 and 3 is that the responses of the human VNO to such substances are not only gender specific, but also species specific. The known pig pheromones, 5α-androst-16-en-3α-ol (androstenol) and 5α-androst-16-en-3-one (androstenone), show no significant activity in the human VNO.

Results: Perfume Components

The next series of experiments was directed more specifically with perfumery in mind. The amplitudes of the EVG responses to estratetraenol, androstadienone, and several

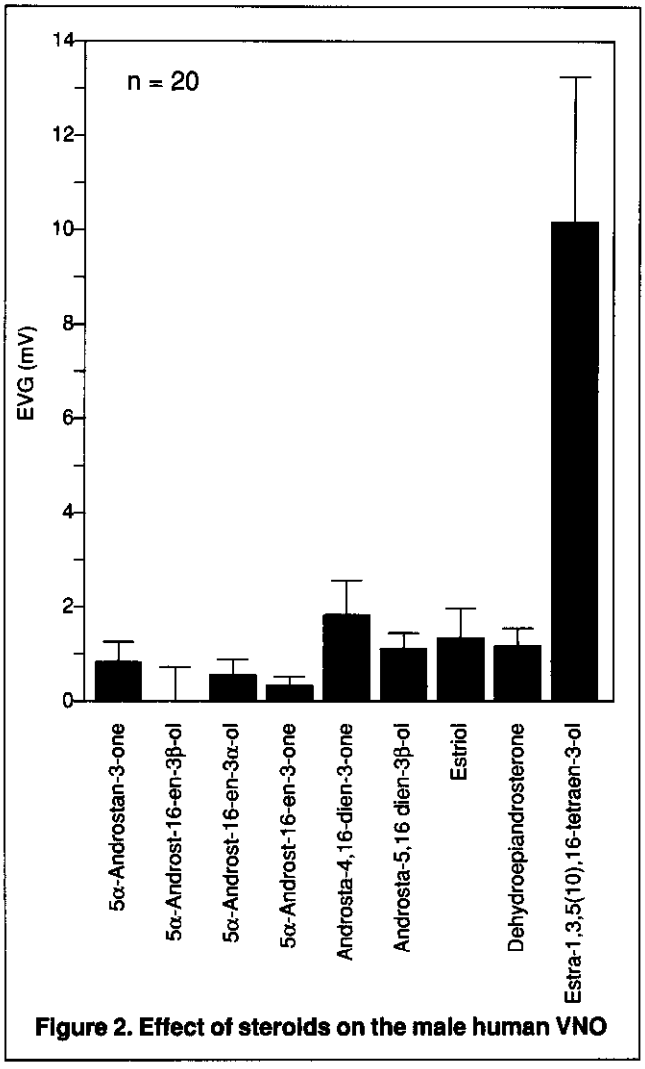

Figure 2. Effect of steroids on the male human VNO

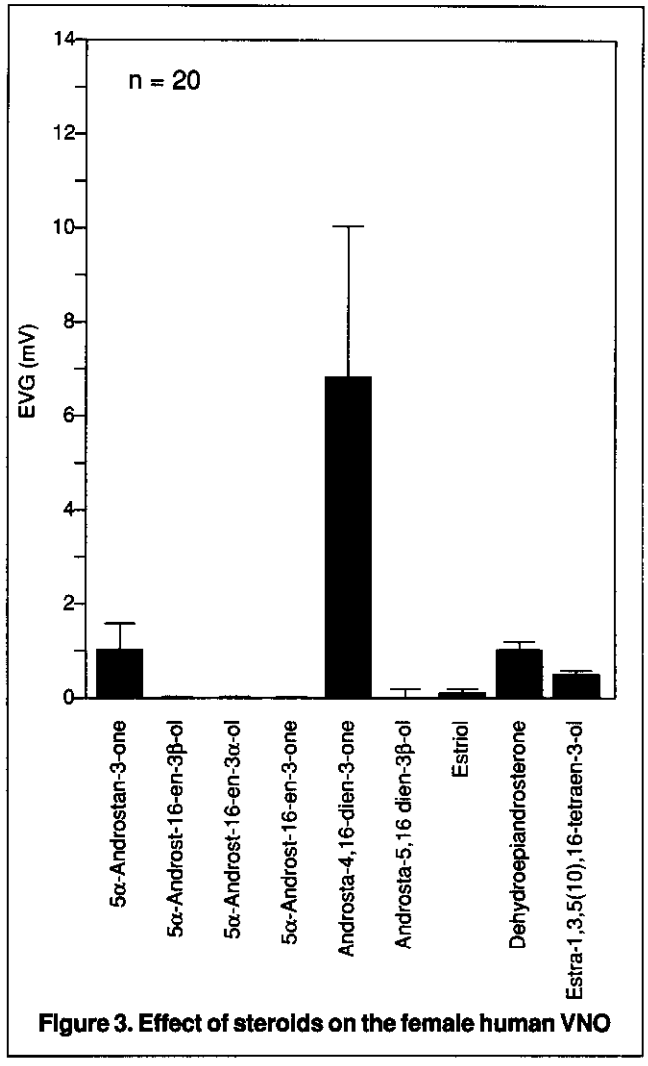

Figure 3. Effect of steroids on the female human VNO

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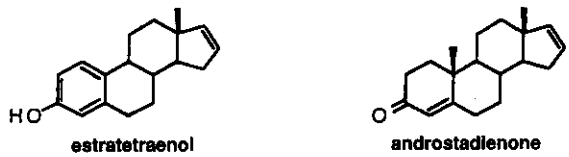

estratetraenol androstadienone

Figure 4. Human pheromones

animal pheromones (civettone, muscone, androstenone, androstenol) and olfactants (tonalid, skatole, l-carvone) of importance in perfumery are shown in Figure 5. Also included is propane-1,2-diol (control), the vehicle in which the other substances are dissolved for testing. Only androstadienone and estratetraenol give rise to significant response.

The pattern of response of the human olfactory epithe-

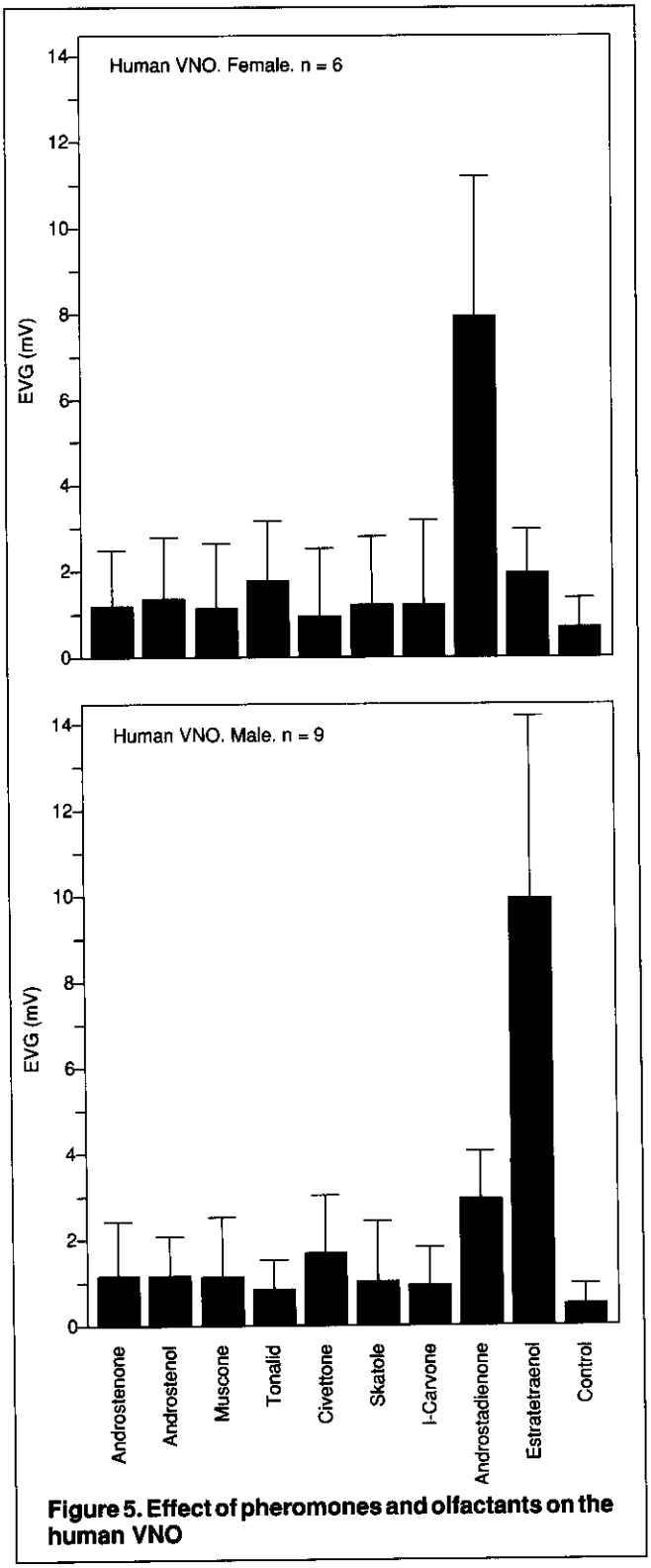

Figure 5. Effect of pheromones and olfactants on the human VNO

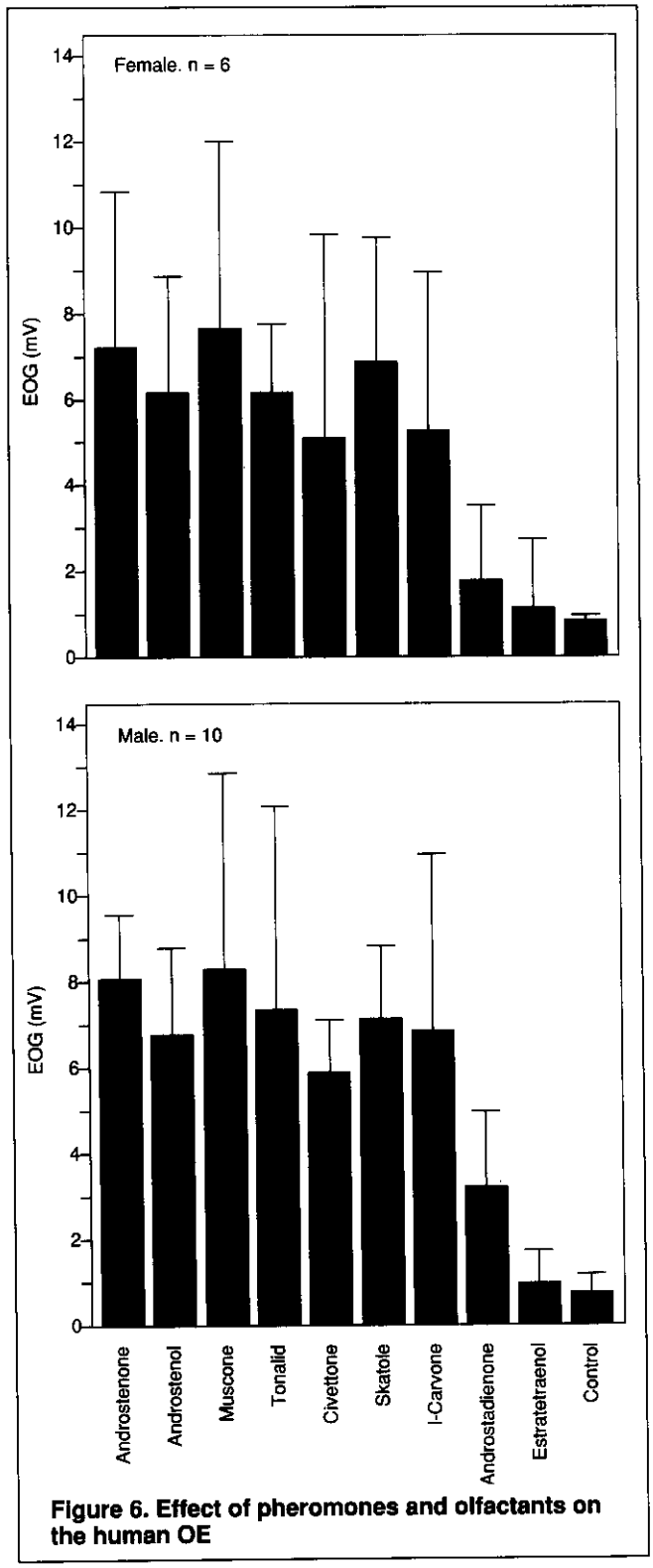

Figure 6. Effect of pheromones and olfactants on the human OE

lium (OE) to pheromones and olfactants (amplitude of electroolfactograms, EOGs), shown in Figure 6, is quite different from the vomeronasal organ response depicted in Figure 5. First of all, olfactants and animal pheromones of importance in perfumery produce a large effect in the OE, whereas the effect of human pheromones is much smaller. This pattern is the exact opposite of what is observed in the VNO. In addition, we observe no gender differences in the response of human olfactory epithelium, again in marked contrast to the vomeronasal system.

Results: Perfumes

The normal use of perfumery products ensures that vapors are automatically delivered not only to the olfactory epithelium but also, by virtue of its location, to the vomeronasal organ. The effective concentration of human pheromones in a perfume will depend on many factors relating to both the nature of the pheromones and the nature of the perfume.

For example, the concentration of pheromone must be sufficient to effectively stimulate the vomeronasal system under the conditions of use. This will depend on both the volatility and the inherent activity of the pheromone. In the case of the particular pheromones under discussion here, the effective concentration is in the micromolar range. Conversely, the concentration must not be so large as to adversely affect the odor qualities of the perfume. Steroidal pheromones are less volatile than the majority of perfume ingredients and therefore act as fixatives in a perfume, regardless of their intrinsic odor.

However, before proceeding with the use of a human pheromone as one of the components of a perfume, it is first necessary to demonstrate that its presence does, in fact, cause VNO stimulation despite the presence of multifarious other ingredients.

Figure 7 depicts the effects of two perfumes on the VNO and OE of women. There are a number of known human pheromones, and the proportions in "Realm" perfumes (Erox) are maintained as a trade secret.[11] However, "Realm for Women" contains, among other components, androstadienone. "Lydia" contains androstenol.[17] Clearly, androstadienone retains its efficacy even when mixed with the other components present in the perfume. The concentration of the pig pheromone androstenol in "Lydia" is five hundred times greater than the concentration of androstadienone in "Realm for Women." Thus the results also demonstrate that it is not possible to compensate for a substance being inherently inactive in the human VNO by increasing the concentration.

A number of commercially available perfumes or perfume additives are claimed to contain pheromones. For the most part, the claimed active component is chosen from the class of androstanes without activity in the human VNO (Figure 8).

In order to correlate the VNO activity of finished products with the activity of their "active" ingredients, some perfumes or perfume additives were tested which are claimed

to contain pheromones. All products were tested "as is" at their commercial strengths, without any dilution prior to testing. The results are shown in Figure 9.

Products containing the pig pheromone androstenol, such as Jovan's "Musk-2," claimed to be "a pheromone-based fragrance," do not exhibit VNO activity in humans because of the species specificity factor. The fragrance additive "Athena Pheromone 10:13" (Athena Institute) is reported to contain dehydroepiandrosterone (Figure 8), which is not active in the human VNO (nor is its acetate or sulfate). "Athena Pheromone 10:13" is also inactive (Figure 9).

In addition, several established perfumes were tested for which no claims regarding human pheromones are made, either explicit or implied. These products do not stimulate the human VNO.

Thus, only products which contain human pheromones are active in the human VNO, and there is excellent correlation between the nature of the components and the activity of the finished products (Figures 2, 3 and 9).

The effects of the products on the olfactory epithelium are shown in Figure 10. As expected, all the effects are large with the exception of "Athena Pheromone 10:13" which contains no odorants other than ethanol.

Discussion

In order to make effective use of human pheromones as components of a perfume, one must first be clear about what is meant by "human pheromones," then establish particular substances which fit the criteria. There has been debate regarding whether the term "pheromone" is ever appropriate for use with mammals, including humans.[18] The term "pheromone" was originally defined in relation to insects,[10] but arbitrary restriction of its use to particular classes of animals diminishes the usefulness of the concept. It seems more productive to recognize that there will be a diversity of responses to pheromones among different animals.

It has been suggested that the word "semiochemical" be used in place of "pheromone" for use with mammals, including humans. However, this idea is confusing and obscures the unambiguous distinction between the two types of substances in humans. The term "semiochemical" includes pheromones, but also includes substances detected only by the olfactory system and eliciting a response which may have a learned component. For example, among skin steroids which are devoid of significant human vomeronasal activity (Figures 2 and 3), there are nevertheless some, androstenone and androstenol, which have an odor. Consequently, they contribute to the olfactory image presented by an individual and so might qualify as semiochemicals, even though they are clearly not human pheromones. Thus, the existence of true pheromones in humans hinges upon the existence and function of the VNO in humans.

The following observations should be taken into consideration when evaluating the status of estratetraenol and androstadienone as human pheromones:

1. The vomeronasal system, which in most terrestrial vertebrates is the principal means for the detection of pheromones, is fully functional in humans. This sixth sense delivers information to specific parts of the brain, just as the other five senses deliver their infor-

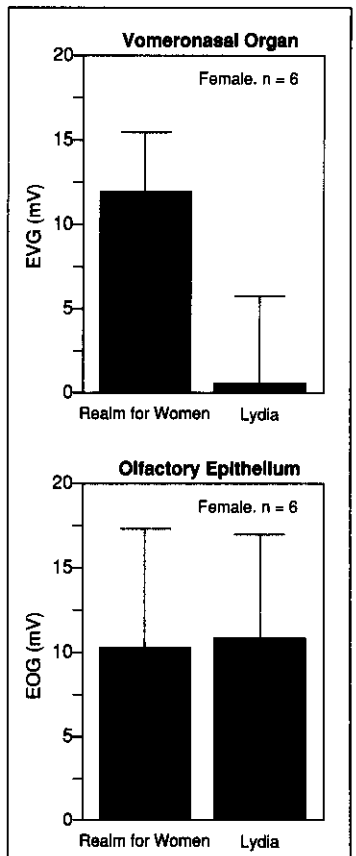

Figure 7. Comparison of the effects of two fragrances

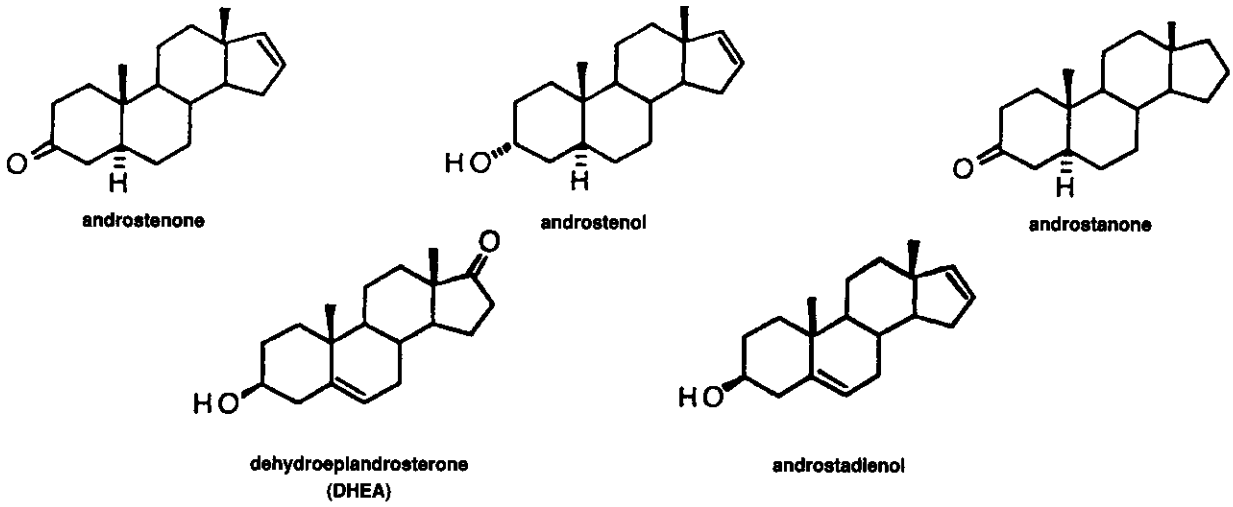

Figure 8. Some androstanes without activity in the human VNO

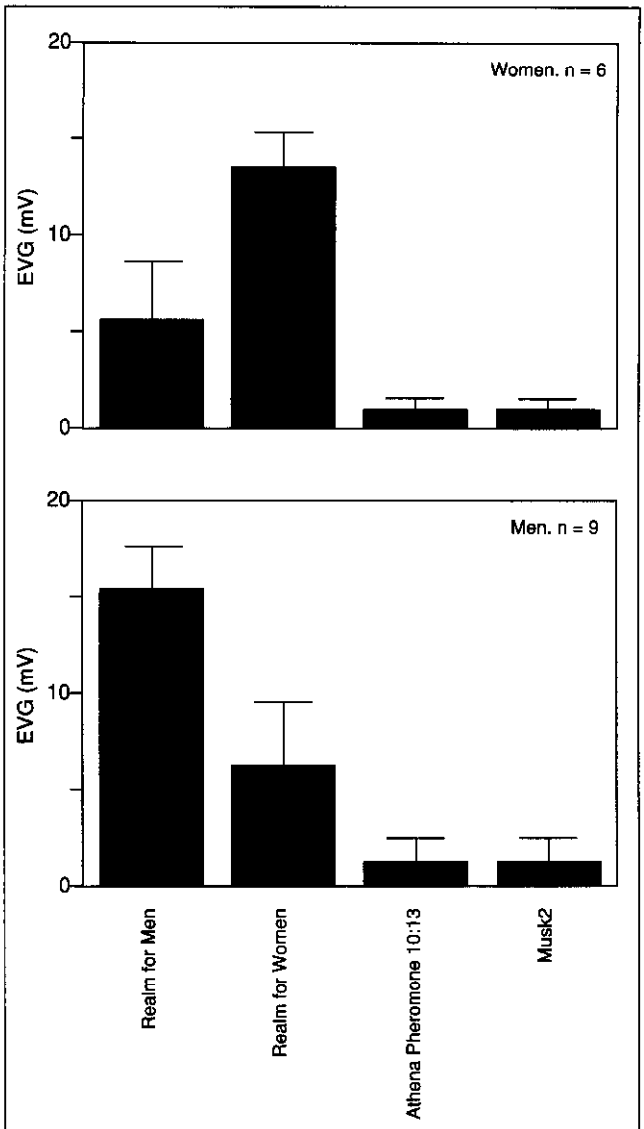

Figure 9. Effect of perfumery products on the human VNO

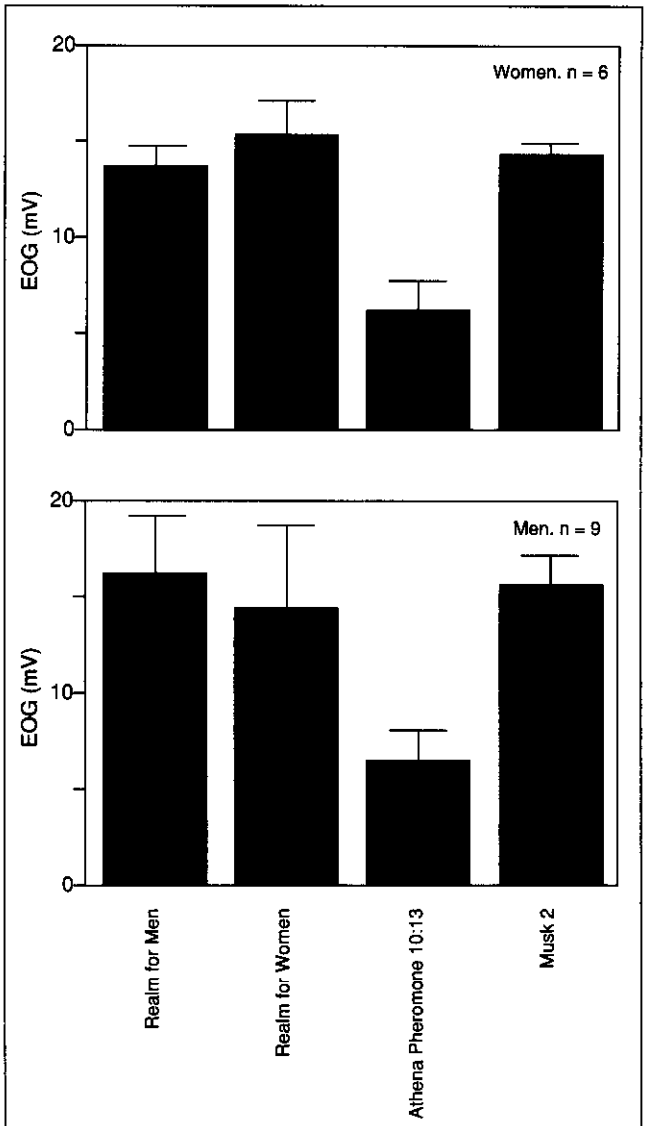

Figure 10. Effect of perfumery products on the human OE

mation to the appropriate part of the brain.[19]

2. Estratetraenol and androstadienone activate the vomeronasal system in humans.
3. I have demonstrated that the closely related pig pheromones, androstenol and androstenone, are not active as pheromones in humans (Figures 2, 3 and 5). Studies of the effect of human pheromones on the VNO of other mammalian species have shown that, at concentrations effective in humans, the effect on the rat VNO is negligible. These data will be published in due course.[20]
4. Behavioral studies of related substances are ongoing. However, my colleagues and I have published elsewhere consistent electroencephalographic effects and electrodermal activity showing clear-cut gender specifity for estratetraenol and androstadienone.[12]
5. The lack of odor of estratetraenol precludes any possiblity of a learned response.
6. The substances under consideration are active in humans as single pure compounds.
7. Testing against control and odorants demonstrates the high specificity of these compounds (Figure 5).

At first sight, it may seem odd that such similar molecules as androstenone and androstadienone would have such dramatically different effects on the female VNO. After all, their olfactory properties are quite similar, as are their electrostatic and hydrophobic binding properties. Indeed, there is evidence that both molecules interact with the same putative olfactory receptor.[13] However, the structure/activity relationships applicable to the VNO receptor for androstadienone are rather different from those applicable to the corresponding olfactory receptor. The lack of activity of androstenone in the human VNO, together with the VNO being sensitive to concentrations of androstadienone below the olfactory threshold, implies that the putative VNO receptor is not merely a transposed olfactory receptor; it is different. Thus, although some human pheromones, such as androstadienone, do have an odor, the olfactory receptor appears to be different from the pheromone receptor in structure, function and anatomic location. No predictions can be made regarding the pheromonal activity of analogs of odorous pheromones based solely on the known structure/activity relationships in the olfactory system.

I have shown that estratetraenol and androstadienone may be used as components in perfumes and can enhance such products. We are approaching a radical shift in the concept of perfumery.[21] Henceforth, the design of a per-

fume should take into account stimulation of the long neglected sixth sense, the vomeronasal system.

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