Review Article

Disease and Smell: “Byoshu”

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Abstract

Following vision, the sense of smell and odor are important sources of external information. However, smell is not acknowledged as an important sensory faculty in clinical medicine and olfaction functions have not been sufficiently examined. Moreover, odor is not used in most diagnoses. Elements of the cause of the change in body odor with aging, *i.e.* aging odors, have been identified. Although it is considered that diseases are characterized by specific odors, particularly cancer, a characteristic known as *byoshu* in Japan, the fundamental nature of *byoshu* has not been clarified. Recently, with developments in techniques for the analysis of odor and greater understanding of the molecular mechanism of the sense of smell, it has become possible to analyze the odor elements that cause *byoshu*. Moreover, the ability to use odor elements and volatile organic compounds to diagnose cancer has also been demonstrated. Development of convenient methods of odor analysis will allow the bedside or outpatient diagnosis of various diseases, including cancer.

KEY WORDS: smell, malodor, volatile organic compounds, cancer, deodorant**Introduction**

The changed odors which occur as an individual ages, in other words in their body odor, are called aging odors. Changes in body odor do not only occur with the physiological processes of aging, or with diet or physical condition, however, but also due to disease.

Disease-specific body odor is useful for medical diagnosis. With the exception of a few specific cases, however, most such changes have not been proven in a scientific manner. Recent improvements in our understanding of the cognitive mechanism of the perception of smell and the development of analysis techniques have prompted a scientific reexamination of this field. In this review, we explain the changes in body odor due to disease, termed *byoshu* in Japanese.

Clinical Aspects of Smell

Medical textbooks have noted that patient odor is useful in the differential diagnosis of disease in comatose patients. Apart from a number of congenital metabolic diseases of infancy, however, body odor due to disease, or *byoshu*, is rarely used as diagnostic evidence in general clinical practice.

Moreover, abnormalities in the sense of smell receive relatively little attention. Harrison's Internal Medicine, a typical textbook of internal medicine, devotes only several pages to descriptions concerning the sense of smell, far fewer than the number devoted to other sensory organs. We therefore consider that the sense of smell and odor has been a relatively overlooked field in recent years.

Smell and Olfactory Sense

Unlike the three primary colors of light, or the five tastes of the taste sense, smell is not comprised of basic essential elements. This lack greatly hampers clarification of the cognitive mechanism of odor and the sense of smell.

The sources of smell are volatile compounds of low molecular weight (molecular weight <300) that are actually recognized as

odor. The estimated total number of odor-producing materials is around several hundred thousand kinds of low molecular weight volatile compounds. Further, it is estimated that 10,000 kinds of low molecular weight volatile compounds are involved in the odor of food, of which only about 5% play a role in imparting smell¹⁾.

Materials that humans can perceive as odor include many organic compounds, such as the aromatic series (benzenes), as well as compounds containing oxygen (alcohol, aldehyde, ketone, and fatty acid), sulfur, and nitrogen, among others. Among inorganic substances, fluorine, chlorine, bromine, sulfur, hydrogen sulfide, nitrogen dioxide, and ammonia, etc., all possess objectionable odors.

Humans perceive odor by inhaling a volatile material into the nose. In general, only about 10% of inhaled material reaches the olfactory tissue²⁾. Odor is not only passively smelled by the nose, but is also actively modified by the brain. Depending on the intensity of an odor, the cerebellum controls the inspiration volume^{3,4)}. Olfactory adaptation decreases sensitivity to strong smells to make it possible to respond to new smells. Bias has a potent influence on the interpretation of smell and behavior derived therefrom. Olfactory recognition is built up by both the nose and brain.

Odor materials such as volatile low molecular weight compounds are recognized by smell receptors that exist in the nasal cavity. Volatile low molecule weight materials originating from other materials or organisms diffuse into air, and thereby reach the nose, where they are dissolved into the mucus of the nasal cavity cuticle in the nasal cavity. Here they bind with olfactory receptors on the microvilli of the olfactory neuronal cells: receptors on olfactory nerves are stimulated, the nerve is excited, and neural impulses are transmitted. The stimulation reaches the cerebral limbic system through the olfactory bulb and tractus olfactorius, enters the olfactory area located in the orbitofrontal cortex, and is recognized as smell.

Olfactory receptors belong to the G protein-coupled receptor family which has seven transmembrane segments⁵⁾ and forms a large family containing catecholamine or angiotensin receptors. With regard to types of olfactory receptor, there are about 1200 kinds in the rat, 1000 in the mouse, 900 in the dog, and 400 in humans (Table 1)⁶⁾. However, although the number of olfactory receptors in humans is less than that in other animals such as dogs, the number of receptor subfamilies is closely similar among species^{7,8)}. It is speculated that the perception of smell is more dependent on the number of receptor subfamilies than on the number of receptors themselves. On this basis, the perception of odor might not differ substantially among animals.

Comparison data between humans and dogs are available. Humans have about 10 million olfactory cells with 8-12 microvilli each, while dogs have about 200 million cells with about 125 microvilli each. These characteristics alone should not be considered to account for the far superior sense of smell of dogs than humans, however; rather, smell is also recognized by the brain, and the difference in how smell is perceived or experienced also results from differences in recognition by the brain. Thus, the superiority or inferiority of the sense of smell should not be judged simply with regard to the number of receptors, number of cells, and sense of smell surface area⁹⁾. As one example, humans and dogs are reported to have no difference in olfactory sensitivity to cocaine (methyl benzoate)¹⁰⁾. The human sense of smell is sharper than might be expected.

Table 1 Numbers of functional genes for olfactory receptors and subfamilies in vertebrate species

	Number of functional olfactory receptor genes	Number of subfamilies
Rat	1201	282
Mouse	1037	241
Dog	872	300
Human	390	299

Byoshu in cancer

The first description of *byoshu* in cancer is a short report in the Lancet in 1989 which described a dog's discovery of a case of malignant melanoma¹¹⁾, and several reports of trained dogs detecting cancer by smell have appeared since. These reports have been heavily criticized, however, on the basis that they did not clarify what the dogs had actually perceived. Improvements in analysis techniques since then now allow the identification of odors and volatile materials in a scientific manner, and diagnostics based on these characteristics has attracted attention.

Basic mechanism of byoshu

Two main mechanisms are considered as generating *byoshu* from cancer.

The first originates from the cancer cells and is derived from the change in the microenvironment. Cancer requires changes in genes to occur. These result in a change in proteins and enzymes, and eventually modified metabolites are produced. These modified metabolites are carried by the blood and excreted from the lungs in expiration, or from the kidneys in urine. They are also secreted from the skin and mucous membranes (especially in the urogenital area). These directly cause *byoshu*. Further, a part of the compounds responsible for *byoshu* is also metabolized by bacterial flora, indirectly causing it (Fig. 1). The type of indigenous bacteria differs by internal organ (Table 2)¹²⁾, and thus metabolism of the material results in different odors, because different bacillus generate different volatile low molecular weight entities. Among such molecules, volatile organic compounds (VOCs) substantially reflect physiological and pathological reactions inside the body and the cell (Table 3)¹³⁾. Such reactions are a phenomenon often seen with cancer cells. It is therefore thought that VOCs will be useful in the diagnosis of cancer.

The second source is odor originating from necrotic organization. Necroses occur in the central portion of cancer tissues that are larger than about 1 cm in diameter. Necrotic organization clearly plays a role in the formation of *byoshu* from cancer. It is well known that the ulceration of breast cancer through the skin is accompanied by a strengthening of body odor.

Fig. 1. Mechanism of the generation of *byoshu* (malodor from disease) in patients with cancer.

Table 2 Typical indigenous microbial flora in the human body

Surface of the body	Head	Gram-positive bacillus Yeast
	Eye	Gram-positive coccus
	Skin	Staphylococcus epidermidis
	Axilla	Coryneform bacterium
Gastrointestinal tract	Oral cavity	Streptococcus
	Stomach	Helicobacter pylori
		Gram-positive bacillus
		Lactic acid bacteria
	Intestine	Streptococcus
		Anaerobic bacteria (ex. Bacteroides) Gram-positive bacillus (ex. E. coli)
Reproductive organs	Vagina	Vaginal lactic acid bacteria

Table 3 Origins of volatile compounds

	Example	Formula	Mechanism of production and diseases
Hydrocarbons			
Saturated	methane	CH ₄	gut bacteria
	ethane	CH ₃ -CH ₃	lipid peroxidation of polyunsaturated fatty acid, oxidative stress
	pentane	CH ₃ -(CH ₂) ₃ -CH ₃	
Unsaturated	isoprene	CH ₂ =C(CH ₃)CH=CH ₂	mevalonic pathway of cholesterol synthesis
Oxygen-containing			
Alcohols	ethanol	CH ₃ -CH ₂ -OH	
Aldehydes	acetaldehyde	CH ₃ -CHO	ethanol metabolism
Ketones	acetone	CH ₃ -CO-CH ₃	decarboxylation of acetoacetate from lypolysis, lipid peroxidation, DM
Carboxylic acids	acetic acid	CH ₃ -COOH	
Sulfur-containing	ethyl mercaptane	CH ₃ -CH ₂ -SH	incomplete metabolism of methionine, elevated in liver impairment
	dimethyl sulfide	CH ₃ -S- CH ₃	
Nitrogen-containing	dimethyl amine	(CH ₃) ₂ -NH	uremia
	anmonia	NH ₄	

Diagnosis of Cancer with Volatile Organic Compounds (VOCs)

Lung Cancer

Recently, investigation has been conducted into the diagnosis of lung cancer by VOCs in expiration. Cancer of the proximal segment of the lung is not easily detected by X-ray examination of the chest. Interestingly, however, cancer cells in this location are often exposed directly to the bronchial tube, highlighting the possibility of diagnosis of lung cancer by volatile elements contained in the expirate. Results to date have been positive compared with those for other cancers¹⁴.

About 3,000 kinds of volatile organic compounds are contained in a human expiration. Of these, however, only about 20 to 30 are commonly noted in all humans¹⁵. VOCs are divided into two groups, namely endogenously produced and exogenous volatiles. Exogenous volatiles include those inhaled from the environment, the oral ingestion of food, and smoking cigarettes. Endogenously produced volatiles include materials newly made from cells in the body and those made by intestinal bacteria. Nevertheless, the physiological meaning and biological origin of many VOCs remains unclear.

Several problems exist in the diagnosis of cancer by VOCs in expiration¹⁶. One is the co-mixing of VOCs originating from the environment. Many volatile low molecular weight compounds occur in our daily environment, and determining whether these are of environmental origin or from inside the body is difficult. The second is expiratory VOC concentration, which is very low, at picomolar levels (10-12mol/L). Measurement of VOCs therefore requires the use of highly sensitive detectors with a technique to concentrate them.

The kind of VOCs included in the expirate of lung cancer patients differs among studies. Some researchers have reported finding lung cancer-specific materials, while others reported only quantitative differences, and a consensus of opinion has not necessarily been obtained. Nevertheless, several VOCs have been commonly noted, and ongoing investigation is anticipated.

Bajtarevic *et al.* analyzed volatile materials of environmental origin in detail (Table 4) and excluded them from the breath analysis of VOCs¹⁷. Based on the 21 kinds of VOCs thereby obtained, sensitivity for the diagnosis of lung cancer was 80%, and specificity was 100% (Table 5).

With regard to the origin of expiratory VOCs, it is not clear whether these are exhausted directly from lung cancer cells to the airway or whether they are first converted to metabolites, enter the blood circulation and are then exhausted via the pulmonary circulation. In studies using cultured cells¹⁸⁻²⁰ and *ex vivo*²¹, detected VOCs were at least partly the same as those from the expiration of lung cancer patients. These findings clearly show the existence of lung cancer cell-specific VOCs.

Breast Cancer

Expiration of breast cancer patients has also been examined. On comparison with VOCs from patients with lung cancer, both the same and different VOCs in expiration have been noted^{22,23}.

Moreover, it has been reported that VOCs are released directly from the skin in breast cancer. Recently, Shirasu *et al.* identified the malodorous component from the gauze of patients with breast cancer and head and neck cancer²⁴. The odor of this element, identified as dimethyl trisulfide, is similar to that of pickled vegetables. They have identified dimethyl trisulfide from different types of cancerous tissue, and consider it to be produced by bacillus in the skin and cancer tissue.

Future development

Diagnosis

Because it is noninvasive and not onerous on patients, diagnosis by VOCs from expiration and the skin has attracted keen interest. Two problems remain to be solved, however, before VOC diagnosis can be established as a part of general diagnostics.

First, VOCs, or the pattern of VOCs, specific to cancer or to cancer types have yet to be identified. More broadly, the mechanism of their production has not been established. Diagnostic use is likely to be considered only after these conditions are clarified.

Second, it is necessary to develop a convenient apparatus for the measurement of VOCs. VOCs are presently measured using GC-MS (gas chromatography-mass spectrometry) after concentrating of the expiration element. This method is well established at the laboratory level, and shows excellent reproducibility. However, the GC-MS equipment is expensive and large, and requires complicated techniques for measurement. Simple and repeated measurement of expiratory air or VOCs is not possible at the medical clinic level, highlighting the need for a small, handy-type instrument. Recently, small-type sensors have been developed, such as colorimetric sensors¹³ and sensors using gold nanoparticles²⁵.

Deodorants

Research into odor and volatile low molecular weight compounds not perceived as odor is ongoing. In the meantime, however, many patients and their families continue to suffer from *byoshu*, and the elements that cause it, even though not identified, should be countered with deodorants. Deodorant methods for malodors like *byoshu* includes physical, chemical, biological, or sensory types (Table 6).

Air cleaner is often used as a physical method. However, the elements and strength of *byoshu* differ from those of odors in the home. A sufficient deodorant effect is not achieved with ordinary air cleaners for family use; rather, improved air cleaners are required, for example by developing stronger filters that are appropriate for *byoshu*, or by introducing new methods of deodorization.

Another physical method already used for *byoshu* is the absorption method with activated carbon. Although cheap and easy to use, its effects have been described as not sufficiently strong.

For *byoshu*, we are using a deodorant material called DEOATAK® (Takasago International Corp., Hiratsuka-city, Kanagawa)²⁶. This material is of plant origin, which uses the deodorant principle of chemically uniting odor materials. Compared with activated carbon, this material has a greater deodorant effect, particularly on nitrogenous substances or sulfated compounds. An effect on *byoshu* is also expected.

Antimicrobial agents are used as a biological deodorant method against *byoshu* associated with bacterial infection. Metronidazoles have been used in many cases. The use of antimicrobial agents has problems, however, such as the side effects of the drugs and the emergence of drug-resistant strains of bacteria.

Another sensory deodorant method is masking, and a commercially available aromatic substance is often used for *byoshu*. However, this approach often produces a more objectionable odor, perhaps due to the mixing of odors. Masking should therefore only be conducted after the completion of sufficient deodorization to reduce *byoshu*.

Table 4 List of compounds whose origins are considered exogenous

Origin			Structure
Hydrocarbons			
Saturated	1,1-Difluoro ethane	Used as a refrigerant, hence an exogenous origin is possible	
	Isobutane	Exogenous origin? (propellant)	
	N-pentane	Marker for oxidative stress	
	Pentane, 2-methyl-	Potentially interesting compound, but might be released by GCMS septa	
	Pentane, 3-methyl	Potentially interesting compound, but might be released by GCMS septa	
	N-hexane	N-hexane pollution in the environment is ubiquitous	
Unsaturated	Ethylene, tetrachloro-	Used in dry cleaning, hence an exogenous origin is possible	
	Ethylene, trichloro-	TCE; groundwater contamination by TCE is an important environmental concern, hence an exogenous origin is possible	
Aromatic	Limonene	Exogenous origin? (is used in food manufacturing, cosmetics and cleansing agents)	
	Benzene, ethyl-	Potentially interesting compound, but one of the volatile BTEX-compounds (= benzene, toluene, ethyl-benzene, xylene) appearing in gasoline	
	Styrene	Styrene is sometimes added to the BTEX-compounds (see ethyl-benzene above), making it BTEXS	
	P-xylene	Indoor air component in hospital rooms	
	P-cymene	Is contained in essential oils (e.g., in cumin and thyme)	
Oxygen-containing			
Alcohols	Ethanol	Could be of exogenous origin	
	2-Propanol	Indoor air component in hospital rooms	
	2-Propanol, 1,1,1-trichloro-2-methyl-	Exogenous origin?	
	Menthol mix of isomers	Might be contained in candies, toothpaste or foodstuff	
Carboxylic acids	Acetic anhydride	Origin unclear	
	Acetyl bromide	Origin unclear	
Esters	Methyl acetate	Observed in healthy volunteers in low concentration (ca. 1 ppb), increases with increased cardiac output	
Ethers	Diethyl ether	Suspected to be an indoor air component in hospital rooms	
	Cineole	Used in flavorings, fragrances, and cosmetics	
Sulfur-containing	Carbon disulfide	Released by GCMS septa	
Nitrogen-containing			
	Acetamide, N,N-dimethyl-	Released by Tedlar® bags	
	Formamide, N,N -dimethyl-	Suspected to be an indoor air component in hospital rooms	

Modified from Bajtarevic A *et al* ¹⁷⁾.

Table 5 Potential breath marker compounds for lung cancer

		Formula	Structure
Hydrocarbons			
Saturated	Butane, 2-methyl-	C ₅ H ₁₂
	N-undecane	C ₁₁ H ₂₄
	Undecane, 3,7-dimethyl-	C ₁₃ H ₂₈
Unsaturated	2-Butene, 2-methyl-	C ₅ H ₁₀
	1-Cyclopentene	C ₅ H ₈
	1,3-Cyclopentadiene, 1-methyl-	C ₆ H ₈
Aromatic	Benzene, cyclobutyl-	C ₁₀ H ₁₂
Oxygen-containing			
Alcohols	1-Propanol	C ₃ H ₈ O
	3-Butyn-2-ol	C ₄ H ₆ O
	2-Butanol, 2,3-dimethyl-	C ₆ H ₁₄ O
Aldehydes	Benzaldehyde	C ₇ H ₆ O
	N-pentanal	C ₅ H ₁₀ O
Ketones	2-Butanone	C ₄ H ₈ O
	2,3-Butanedione	C ₄ H ₆ O ₂
	2-Butanone, 3-hydroxy	C ₄ H ₈ O ₂
	Acetophenone	C ₈ H ₈ O
Esters	Butyl acetate	C ₆ H ₁₂ O ₂
Sulfur-containing			
	Methyl propyl sulfide	C ₄ H ₁₀ S
Nitrogen-containing			
	Urea, tetramethyl-	C ₅ H ₁₂ N ₂ O
	Ethylenimine	C ₂ H ₅ N
	Isoquinoline, 1,2,3,4-tetrahydro-	C ₉ H ₁₁ N

Modified from Bajtarevic A *et al* ⁽¹⁷⁾.**Table 6** Methods for eliminating odor

1. Physical	Aqueous cleaning Air ventilation Absorption to activated charcoal
2. Chemical	Combustion Neutralization • oxidation Chemical absorption
3. Biological	Microorganism Antimicrobial agent
4. Sensory	Masking with air freshener

In Conclusion

With ongoing development of analysis methods, it is expected that diagnosis with volatile materials will become possible not only for cancer but also various other diseases.

The belief that a change in body odor is simply due to aging might result in overlooking the possibility that the change is due to a hidden sickness. The establishment of diagnostics by odor may facilitate the speed and convenience of diagnosis, thus likely leading to the earlier detection and treatment of disease.

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