

The Chemistry Decomposition in Human Corpses

BEATRICE GABRIELA IOAN^{1,2}, CRISTIANA MANEA³, BIANCA HANGANU^{1,2*}, LAURA STATESCU^{1*}, LAURA GHEUCA SOLOVASTRU¹, IRINA MANOILESCU^{1,2}

¹Grigore T. Popa University of Medicine and Pharmacy, 16 Universitatii Str., 700115, Iasi, Romania

²Institute of Forensic Medicine, 4 Bunavestire Str., 700455, Iasi, Romania

³Alexandru Ioan Cuza University of Iasi, Faculty of Geography and Geology, 22 Carol I Blvd., 700506 Iasi, Romania

Human body is a complex of organic substances (proteins, lipids, carbohydrates), which undergo chemical decomposition processes soon after death. The compounds released during decomposition characterize the development of different stages of this process: e.g. biogenic amines resulted from the proteins decomposition will confer the particular smell of a cadaver; gases resulted from carbohydrates fermentation will give the bloating aspect of the cadaver. The study of cadaver decomposition and the products resulted from this process is the subject of human taphonomy and is realized nowadays in special facilities in USA and Australia. Identification and analysis of the chemical compounds emerged after human decomposition (gases, liquids, salts) give valuable information to forensic pathologists for estimating the postmortem interval (PMI). More, volatile compounds – which give the odor signature” specific to human remains – may be utilized in identifying clandestine burials, human remains or victims entrapped under ruins in cases of natural disasters. In this paper the authors describe the chemical decomposition stages of human cadavers, the factors influencing these processes and utility for the forensic activity of the results of human taphonomic studies.

Keywords: decomposition compounds, chemical processes, postmortem interval, human taphonomy, forensic medicine

After death, the human body undergoes the decomposition process and its constituents: 64% water, 20% proteins, 10% lipids, 1% carbohydrates and 5% minerals [1] are broken down into simpler compounds, until they reach their building block ingredients, i.e. C, H, O, N, P and S [2]. These events are studied during what we call the chemical decomposition of corpses (postmortem decomposition).

The postmortem decomposition evolves in two main phases: the first one is autolysis which consists of the enzymatic self-digestion of the cells [3] and paves the way for the second phase, i.e. putrefaction. The trigger for these processes is the cell anoxia- cessation of oxygen delivery to the cells- which intervene between the cell membranes and junctions destroying them, the enzymes and the entire content of the cells being released [4, 5]. Lack of oxygen also creates optimal conditions for the development of anaerobic bacteria in the gastrointestinal and respiratory tracts, such as *Bacteroides* and *Clostridium* [4].

Factors influencing cadaver decomposition

Decomposition process follows the same stages in every corpse, but the particularities of the body and the adjacent environment make it happen in different moments in time (table 1).

Particularities of the body comprise: clothing, age, body mass, body dimensions, gastric content [6], hydration state

[7, 8] and nutritional state before death, as well as cause of death [7]. Regarding the latter, the sepsis- which maintains the body temperature at high levels [5, 9], asphyxia- when the blood remains in a liquid phase favoring the spread of the bacteria [10] or congestive heart failure [7] - which confers a high level of internal hydration by edema, hasten the process of putrefaction.

Characteristics of the environment in which the body stays [4] and which are relevant for the development of the chemical decomposition are: temperature, humidity, pH and composition of the soil in case of buried corpses [6, 8, 11]. Decomposition in corpses sitting outdoors is significantly influenced by the activity of insects, rodent and carnivores [2, 10, 11] and the stream of air [2, 7], as well as the rainfall [11].

Humidity in the environment and inside the body (antemortem hydration, congestive heart failure) hastens the putrefaction process [7]. Decomposition is also speeded up by obesity and warm clothes. On the contrary, this process is delayed by tight clothes and placing the corpse on metallic surface [9].

Temperature and humidity also influence the profile of the volatile compounds released during the putrefaction process in two ways: first, by the direct effect they have on the breakdown of proteins and carbohydrates and second by influencing the type of microorganisms in the soil [2, 6] and insects which colonize the body [11].

Particularities of the body	Particularities of the environment
Clothing	Temperature
Age	Humidity
Build	Soil composition
Gastric content	Soil pH
Diseases/pathologic state	Stream of air
Cause of death	Insects, microorganisms

Table 1
FACTORS AFFECTING THE RATE OF CADAVERIC
CHEMICAL DECOMPOSITION

* email: bianca_h_no1@yahoo.com, laura.statescu@umfiasi.ro

The profile of the volatile compounds generated during the chemical decomposition is also influenced by different species of bacteria, such as *C. aminovalericum* which produces acetone, ethanol and 1-butanol, and *C. cadaveris* which produces in addition isobutanol, *n*-butanol, isoamyl alcohol and *n*-amyl alcohol [2]. The presence of larvae emerged after scavenger insects oviposition contributes in addition to the destruction of the tissues [4]. Low temperature will gather less insects, which will destroy less cadaveric material, leaving more tissue for chemical conversion by bacterial activity, leading further on to successive leakage of the fatty volatile acids in the soil [11].

The fact that human postmortem decomposition is a process and not a solitary event is proved by the release of several volatile compounds in certain PMIs [12], or by the presence of multiple stages of decomposition on the same corpse at the same time [13].

Putrefaction, both transformative and destructive process [10], implies progressive disintegration of the organic compounds [10] from the tissue to form gases, liquids and salts [5, 7]. Unlike autolysis, putrefaction is noticeable on the external examination of the body since its beginning by different aspects such as color changes and peculiar odors, and bloating of the body [3, 4], the ground for these changes being prepared both by autolysis and by different microorganisms: bacteria, fungi and protozoa [2].

The release of different volatile compounds during putrefaction determines specific odors: ammonia has pungent smell, hydrogen sulfide smells like rotten eggs, sulfur dioxide has an irritating and pungent odor, indole and skatole – fecal and nauseating odor, cadaverine – putrid and decaying flesh odor, putrescine- putrid and nauseating odor [2].

Changes during the first stages of cadaveric chemical decomposition cannot be seen with the naked eye [5, 14], but the microscope examination reveals the gradual fainting of the cells and tissues structure [14]. Corpses spread out particular odors since the very first minutes after death [15]. These initial odors cannot be perceived by human senses, but the flies have the capacity to sense them due to their antennas with special functions [2]. Thus, in the early stage of decomposition, flies will populate the corpse and lay eggs especially around the orifices [16].

The first sign of putrefaction at gross examination is the greenish coloration of the skin in the right iliac fossa [7, 16]. The specific location is due to the close contact between the caecum and the skin at this level [7, 16] while the peculiar color is given by sulfhemoglobin, arisen from the hemolysed hemoglobin coupled with the hydrogen sulfide generated by bacteria from the intestines [14]. Combining the enzymatic action during the breakdown of erythrocytes (ensuing hemoglobin and then iron) with the bacterial action producing hydrogen sulfide, a dark color residue will form [14], which will be deposited on the blood

vessel walls and subsequently in the tissues, giving the mottled aspect of the skin [14].

During the chemical process of postmortem decomposition a number of gases will evolve (CO_2 , CH_4 , NH_3 , H_2S , SO_2 and H_2), resulting in the distention of the tissues [2, 5]. Further on, the pressure inside the body will increase and the purging fluid will be expelled, first through oral and nasal cavities and then through every opening of the body [4, 7]. The distension of the body in this stage may determine ruptures of the skin, and together with the destructions resulting from larval activity, the tissues will be receiving new oxygen from the environment, and the activity of the aerobic microorganisms will be resumed [4]. Withal, the gases evolving inside the abdominal cavity will elevate the diaphragm, increasing pressure inside the thoracic cavity and as a consequence the air will mix the putrefaction fluid with the expulsion of reddish foam from nose and mouth [16].

Cadaveric decomposition ends with skeletonization, a phase which sometime needs days or months and sometimes years to be reached [14] while the complete disappearance of the corpse may need hundreds of years [14].

The metabolism of the aerobic bacteria which are normally present inside digestive tract and respiratory system exhausts the minimum oxygen remaining in the body following cessation of the cardiac activity [17, 18], and the ground will become propitious for anaerobic organisms, such as *Bacteroides*, *Clostridia*, *Streptococci* species which will enable the conversion of proteins, carbohydrates and lipids to different organic acids and gases [4, 14].

Protein decomposition

During the chemical postmortem decomposition, proteins are first broken down mainly to proteoses, peptones, polypeptides and amino acids [4, 14, 18] through proteolysis, by the action of enzymes [14]. Further on, proteins are reduced to nitrogen, phosphorus and sulfur compounds [2], the latter being represented mainly by dimethylsulfide [12]. Intermediate products to evolve from this process are biogenic amines which are by-products of proteolysis [14]. The most important biogenic amines are histamine, putrescine (resulted from L-ornithine decarboxylation), cadaverine (resulted from L-lysine decarboxylation), tyramine, tryptamine, beta-phenylethylamine, spermine and spermidine [2, 13]. Among these, histamine, tryptamine and phenylethylamine are accompanied by the evolution of gases such as methane and carbon dioxide [14], while the toxic diamines (cadaverine and putrescine) confer the characteristic smell of decomposing bodies [14] (fig. 1).

Muscle tissue proteins, membrane proteins and free proteins consist mainly from amino acids [13]. Some amino acids contain sulfur atoms and during the desulfhydrylation process they may be reduced to form

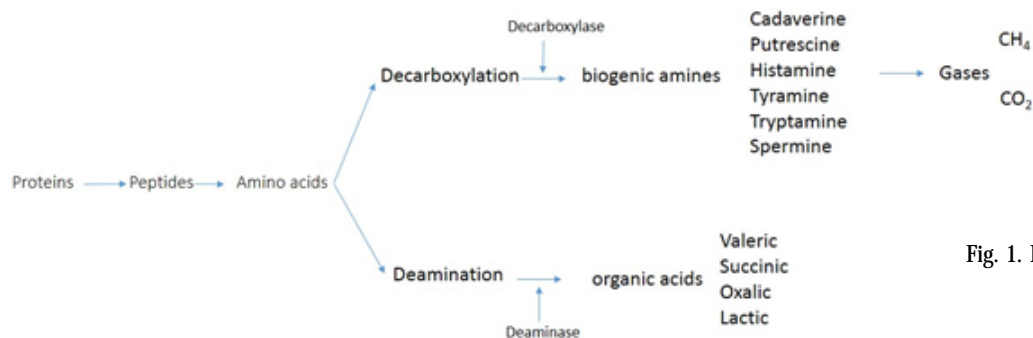


Fig. 1. Postmortem decomposition of proteins

ammonia, thiols, pyruvic acid, hydrogen sulfide and sulfides [14], the latter being favored by the soil anaerobic conditions for the buried corpses [14]. In burial sites, the usually anaerobic environment from the ground stops the process in this stage, and the sulfides are not transformed further [14, 18]. Instead, oxygen in aerobic conditions will assist the transformation of sulfide in sulfate, and specific bacteria (e.g. *Thiobacillus*) will contribute to their further transformation in sulfurous acid [14, 18] (fig. 2).

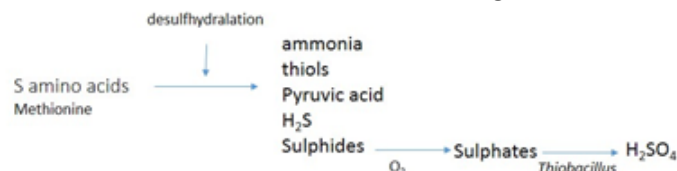


Fig. 2. Postmortem decomposition of sulfur amino acids

During the same process of decomposition, nitrogen from amino acids is released in the form of ammonia (NH_3), and the acidic conditions in the soil (in burial environments) will convert the ammonia to ammonium (NH_4^+). The rest of ammonium will follow two ways: nitrification and denitrification [14, 18], in both processes being involved different bacteria. During the nitrification process [aerobic], ammonia is transformed in nitrite and further on in nitrate. Denitrification (anaerobic) induces the reduction of nitrate to nitrite then to gaseous nitrogen and nitrous oxide [14, 18] (fig. 3).

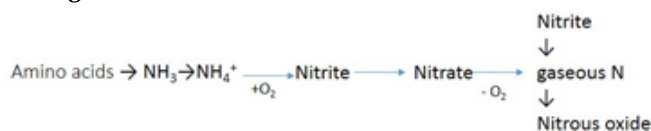


Fig. 3. Nitrification and denitrification process

The rate of decomposition of proteins is not the same in every tissue. Neuron membrane proteins and proteins from the epithelial tissue of the gastro-intestinal tract are the first to be decomposed, while reticulin, collagen, epidermis and muscle proteins are much more resistant to putrefaction [14, 18]. Keratin, the protein from the cornified layer of the epidermis, is resistant to most proteolytic enzymes [4, 18], this being an explanation for persistence for longer periods of time [18]. This resistance owes to disulfide bonds in the structure of cysteine [18]. Sulfides in the skin will undergo desulfhydrylation in order to be transformed into sulfuric compounds such as dimethylsulfide and thiol [19].

Carbohydrates decomposition

From the postmortem decomposition of the carbohydrates will result mainly oxygenated compounds such as: alcohols, aldehydes, ketones, acids, ethers and esters [2, 20]. In the first stage of this process the microorganisms will convert glycogen to glucose monomers. Glucose will follow afterwards two ways: complete decomposition by oxidation, leading to formation of CO_2 and water [13, 14, 18] or incomplete decomposition, leading to the evolution of several organic acids (citric, oxalic or glucuronic acids) [13] and alcohols [14, 18],

causing an acidic environment around the body (fig. 4). In anaerobic environment, different microorganisms will be implicated in acids and alcohol formation: bacteria will assist the formation of lactic, butyric and acetic acids [14, 18, 21], as well as ethanol and butanol formation [13, 21]. Instead, aerobic conditions will enable fungi to assist the formation of glucuronic, citric and oxalic acids [13, 14, 21]. Bacterial fermentation will produce methane, hydrogen sulfide and hydrogen gas [18, 21]. When muscular activity was recorded prior to death, the glycogen in the muscles will be decomposed to lactic acid earlier than in case of muscle rest, as a consequence of precocious anaerobic environment formation [13].

Several bacteria mediate carbohydrates decomposition. *Clostridium* ferments the pyruvate from carbohydrates to acetone, ethanol, acetic acid, butanoic acid [22]. *Enterobacteriaceae*, and particularly *E. coli* ferments the pyruvate to lactic acid, succinic acid, acetic acid, formic acid and ethanol [22]. *Streptococcus*, a facultative anaerobic bacterium, ferments carbohydrates to lactic acid [13]. *Clostridium perfringens* has saccharolytic, proteolytic and lipolytic abilities [18].

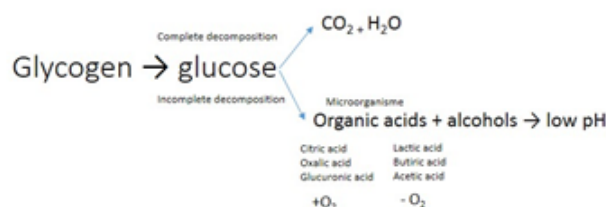


Fig. 4. Postmortem decomposition of carbohydrates

Lipids decomposition

Fatty tissue consists, on the average, of 5-30% water, 2-3% proteins and 60-85% lipids, of which 90-99% are triglycerides. The latter are made of one glycerol molecule attached to three molecules of fatty acid [23]. Cellular membranes consist of phospholipids and their break down is accomplished by phospholipases [13, 24].

Lipids will be decomposed in hydrocarbons, nitrogen, phosphorus and oxygenated compounds [2, 20], palmitic acid and oleic acid [20]. Initially, lipids are transformed to triglycerides by the action of lipases [21]. Triglycerides will next form saturated and unsaturated fatty acids by hydrolysis and by lipases activity.

Further, the characteristics of the environment guide the decomposition. Thus, in aerobic conditions the unsaturated fatty acids are oxidized to odoriferous aldehydes and ketones [4, 21], while anaerobic conditions will favor hydrogenation of the unsaturated fatty acids, to form saturated fatty acids, compacted as a solid mass called adipocere [18, 21].

Some of the fatty acids are volatile, such as propionic, valeric and butyric acids [21] (fig. 5).

Acetoacetate resulted either in the process of lipids peroxidation or lipolysis will be decarboxylated in order to form acetone [2].

Polyunsaturated fatty acids compounding cellular membrane will be converted to volatile alkanes by oxygen free radicals during lipid peroxidation process [12]. The

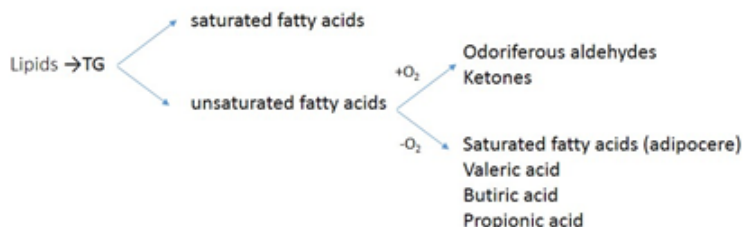


Fig. 5. Postmortem decomposition of lipids

first to be converted are long chain fatty acids, which will be split in smaller fatty acids and afterwards a new hydrolysis will take place in order to produce oxy-fatty acids [18]. On the one hand, the lower pH (as a consequence of lactic acid accumulation) stops the activity of lipoprotein lipase and on the other hand upholds the activity of lysosomal acid lipase [13].

Relevance of the human taphonomy studies for forensic medicine

Numerous substances are produced within the chemical processes of human cadaver decomposition (table 2). Their identification is relevant to the forensic activity [25, 26] as they offer valuable information for the location of clandestine graves, for the detection of the victims entrapped under ruins in case of earthquakes and for the estimation of PMI.

Ammonium resulted following decomposition process of the amino acids from proteins is a rich nutrient for vegetation [14, 18], which makes the soil covering corpses in shallow graves to be early covered with a high amount of vegetation, this being an indicator for the detection of human remains or clandestine graves [14].

Decomposition by-products, i.e. volatile organic compounds [2, 3, 6, 13] have their own role in the detection of clandestine graves [4, 15], of human remains [3] or in the detection of victims following natural disasters (e.g. earthquakes) caught under ruins [12]. Likewise, they may be helpful in the manufacture of devices which can detect cadavers in the situations mentioned above [15] or in the training of human remains detection dogs, when the compounds released by decomposing cadavers may be used to test their alerting response [8].

The understanding of chemical postmortem changes is a crucial step in the estimation of the PMI [5]. Still, establishing the PMI is less accurate as decomposition progresses [5], thus being necessary to corroborate as many parameters as possible.

The volatile organic compounds attract different insects [12, 15] which have adapted their olfaction over the years of their development in order to detect specific cadaveric volatile compounds [13] and which will thus colonize the

body at different moments in time, being a very valuable help for the forensic entomologists in establishing the PMI [12]. For example, the insects from *Sphaeroceridae*, *Piophilidae*, *Fanniidae* and *Phoridae* species are attracted by rancid fats, evolved during butyric fermentation phase of putrefaction and ammoniacal fermentation with putrid tissue decay, which happens around 3-6 months postmortem [10]. Developing stages of the insect may be useful in establishing PMI, but as the environment conditions influence in different ways larvae development, there is a need for additional indicators, corroborated with information provided by the entomologists [13].

Several authors attempted to estimate the PMI through a detailed analysis of the volatile organic compounds released during different decomposition phases. Thus, some of them investigated the soil around the buried corpses [8] or the soil under the corpses laying on the surface [11], the air around the corpses in closed environments [2] and above-ground, in forest biotope [15]. Such studies were performed either on pig carcasses, which are the analog of human body [15], or on human corpses [8, 27].

Dekeirsschieter et al. [15] analyzed the samples in their study by comprehensive two-dimensional gas chromatography coupled with time-of-flight mass spectrometry (GCxGC-TOFMS) and they identified 832 volatile organic compounds (10 times more than using GC-MS). They considered all the environmental variables (temperature, humidity, wind speed and wind direction) during 6 months of spring. Environment conditions and other circumstantial elements were underlined by Campobasso et al (2001) as a constitutive part of the experiments in order to manage a suitable estimation of the PMI [10]. The most powerful compounds were identified in active decay stage, after which the number of compounds decreased together with the disappearance of soft tissues [15] (table 3).

Vass et al. [8] identified the following volatile organic compounds released by buried corpses: compounds detected throughout the entire process of decomposition in soil, compounds that may be identified even 16 years after death: benzene derivatives, halogen compounds and

Table 2
CHEMICAL SUBSTANCES RESULTED FROM POSTMORTEM DECOMPOSITION OF PROTEINS, CARBOHYDRATES AND LIPIDS

Proteins decomposition	Carbohydrates decomposition	Lipids decomposition
- Proteases - Peptones - Polypeptides - Amino acids - By-products: biogenic amines (cadaverine, putrescine, histamine, tyramine, beta-phenylethylamine, spermine, spermidine)	- Alcohols - Aldehydes - Ketones - Acids - Ethers - Esters	- Hydrocarbons - Nitrogen - Phosphorus - Oxygenated compounds - Palmitic acid - Oleic acid

Table 3
VOLATILE ORGANIC COMPOUNDS RELEASED DURING SPECIFIC DECOMPOSITION STAGE [15]

	Fresh decay (first 5 days)	Bloating stage (next 15 days)	Active decay (next 13 days)	Advanced decay (last 11 days)
Dekeirsschieter et al. (2012)	Mainly alcohols (1-butanol) and alkenes in day 1	Alcohols decreased Ketones increased – mainly aldehydes, aromatic compounds and sulfur compounds	Nitrogen and sulfur compounds	Carboxylic acids decreased Ketones and alkanes increased

Table 4
VOLATILE ORGANIC COMPOUNDS LIBERATED DURING SPECIFIC DECOMPOSITION STAGE [11]

	Fresh decay	Bloating	Active decay
Vass et al. (1992)	Insignificant changes	By-products rich in butyric acid, resulted during anaerobic fermentation in the intestines	Aerobic and anaerobic bacteria by-products, which will disappear by the beginning of the 4 th stage

aldehydes; compounds evolving only in the early stages of decomposition, being detected in the first year after death or shortly after this interval and which tend to be the most cyclic: esters, certain benzene derivatives and some halogen compounds; compounds existing on the surface of the graves as long as the soft tissues of the body persist, including mummified stage: many of the sulfur compounds and of the halogenated compounds. They also mention that in the first 17 days after burial, no volatile organic compounds are detected at the surface of the grave [8].

Vass et al. [11] conducted a study to estimate the PMI by analyzing the soil underneath 7 cadavers (soil solutions), considering soil humidity, environment temperature and the body mass. Volatile fatty acids that may be identified in soil solutions are only those which are hydro soluble: formic, acetic, propionic, butyric, valeric, caproic and heptanoic acids. Based on exclusion criteria (their presence in nature, variation according to season and environment temperature), the most useful in the estimation of the PMI are the propanoic, butyric and valeric acids [11]. Based on sequential decomposition of proteins and carbohydrates the authors succeeded to correlate decomposition stages with the production of volatile fatty acids, this being useful in estimation of the PMI (table 4). However this method is not suitable in case of graves located in areas with frequent floods or with a high level of humidity or in mummified and charred bodies [11].

Conclusions

The chemical processes occurring during the human cadaver decomposition generate numerous chemical compounds, the type and rate of their occurrence depending on several factors, either concerning the cadaver characteristics or the characteristics of the environment the cadaver stays.

The identification of chemical substances resulted during the cadaver decomposition is relevant for the forensic activity, allowing the detection of the clandestine burials, of the victims of natural disasters and the estimation of the PMI.

Given the increased number of factors influencing both quantitatively and qualitatively the dynamics of the cadaveric chemical decomposition, there is a need for extensive studies regarding these processes, studies which need to consider as many variables as possible. However, human taphonomy studies which require particular conditions, in specially arranged facilities, which can reproduce as precisely as possible different conditions in which decomposition of human bodies may take place.

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