Shape changes of ripening mistletoe berries

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Translation of: Flückiger, H. and Baumgartner, S. (2002): Formveränderungen reifender Mistelbeeren. *Elemente der Naturwissenschaft* **77** (2), 2-15.

Summary

Path curves are fundamental constructions of projective geometry. They might also serve as single-parametric archetypal forms in the plant and animal kingdom. This hypothesis is based on the observation that a large variety of buds, cones and eggs show a striking similarity to path curves. The outline of mistletoe (*Viscum album* L.) berries also follows a path curve geometry.

We investigated temporal changes in the shape of mistletoe berries by means of empirical determination of the corresponding path curve form parameter λ . Periods of observation reported here were several months in the years 1995, 1997 and 1998.

The existing hypothesis of a correlation of the form parameter λ with alignments of the moon with other planets was falsified. Correlation with various meteorological factors or with solar activity was also not found.

In contrast, a tentative hypothesis of a correlation between the shape of mistletoe berries and the position of the moon in the zodiac, based on the results of the year 1995, was verified in two successive years (1997 and 1998). It seems as if the λ value increases if the moon is situated in the astronomical zodiac signs Gemini, Cancer, Libra, Scorpio, Aquarius and Pisces ('Air' and 'Water' signs). Correspondingly, a position in the signs of Aries, Taurus, Leo, Virgo, Sagittarius and Capricorn ('Fire' and 'Earth' signs) is correlated with a decrease of the λ value. The results warrant further investigations.

Introduction

Egg shapes as mathematically defined path curve surfaces (Ostheimer & Ziegler 1996) can be found not only in the animal kingdom and the human organism but also in the plant kingdom. For instance, leaf and flower buds exhibit such shapes, but fruit nodes are not as a rule egg shaped in the way meant here, even if they are oval (Edwards 1982).

The form of these egg shapes is exactly determined by the single parameter λ (Edwards 1982). $\lambda=1$ gives rise to rotational ellipsoids, $1<\lambda<\infty$ to egg shapes pointing upwards and $0<\lambda<1$ to egg shapes pointing downwards. Resting buds generally have λ values >1 and at bud opening change to <1.

In his fundamental work *The Field of Form*, Edwards (1982) described not only the mathematical derivation of path curve surfaces but also practical procedures for their measurement based on numerous examples. For decades he collected measurement data on a wide variety of plant buds which are determinable by the shape parameter λ . During this work he observed changes in shape of resting buds which correlated with cosmic rhythms. In particular they showed a connection with moon and planetary conjunctions and oppositions. Interestingly, in the case of tree buds, attention was given to the planets

which are traditionally assigned to their corresponding trees. So for instance oak buds showed rhythmic changes that correlated with moon-mars conjunctions and oppositions (Edwards 1992-98).

Defining the problem

A focus for work at the Society for Cancer Research (Verein für Krebsforschung, Arlesheim, Switzerland) involves the investigation of mistletoe plants (*Viscum album* L.) in connection with their host trees. With a view to identifying especially favourable times for harvesting mistletoe for the manufacture of the cancer remedy *Iscador*, it was interesting to discover whether mistletoe was aligned according to the rhythm of the host tree or was emancipated from it and following its own rhythm.

Sonder (1993) documented the λ value of mistletoe buds and berries over a period of 15 months in a weekly rhythm. Whereas the determination of the shape of mistletoe buds requires a greater technical effort (photography of sections) the shape of berries can be obtained with greater ease by photocopying them (see below). For this reason we have here concentrated on studying the berries.

In initial experiments (1990-1) it was possible to confirm that ripening mistletoe berries of various host trees (apple, oak) conform to path curve surfaces. But the fortnightly rhythmic shape changes expected on the basis of the results of Edwards (1982) were not detected. Nevertheless, as the λ value variability on different days was greater than expected given the precision of measurement, we suspected unknown influencing factors and carried out further series of measurements which are here described in greater detail.

Methods

Harvesting berries Several shoots yielding 15-20 berries on each harvest day were picked from previously marked mistletoe clumps on the same small apple tree twice or thrice weekly between 8 and 9 am on each occasion in the years 1995, 1997 and 1998. The



Figure 1.

Tangents and cross axis of a mistletoe berry for determination of the λ value

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berries sampled each time had an average degree of ripeness.

In 1995 the period of investigation began in July with the as yet green berries and ended in December when they were fully ripe. In 1997 and 1998 the sample period was restricted to October to December.

Immediately after harvest each berry together with a small piece of stem was carefully cut off the shoot without damage, otherwise the shape can be measurably changed. Each berry was photocopied in two or three different positions (enlarged twofold). With ripe berries a protective ring was used to minimise shadows on the copy and deformation of the berries by the lid of the photocopier.

Measurement of berry shapes and calculation of λ values Photocopies of the berries were selected that were sufficiently sharply outlined and were not tilted with respect to the light source. This is achieved when the stigma at the upper end of the berry coincides exactly with the outline of the circumference (see Fig. 1). Measurement of the berries followed the indications of Edwards (1982). Tangents were placed at the base and tip of the berry image and the long axis drawn in (Figs. 1 & 2). In doing so, the stigma was not treated as belonging to the berry shape. The lower tangent has to be placed with a certain 'sense for form'. The long axis of the berry was divided into eight equal sections, intersecting the berry at various levels (Fig. 2: T, A-F). Level T bisects the long axis. The diameter of each section was measured at each level and related to the diameter at level T (for an example, see Table 1). For each level, λ values were calculated according to the formulae in Table 2.



Figure 2.

Outline of an upwards pointing (λ =1.05, left) and a downwards pointing egg shape (λ =0.95, right).

Level	Diameter	In relation to T
F	9.4 mm	F/T = f = 0.667
E	12.3 mm	E/T = e = 0.872
D	13.7 mm	D/T = d = 0.972
Т	14.1 mm	
С	13.6 mm	C/T = c = 0.965
В	12.1 mm	B/T = b = 0.858
А	9.1 mm	A/T = a = 0.645

Table 1.

Numerical example for calculation of the relative diameter a, b, ..., f. The latter forms the basis for the calculation of λ values (see Table 2)

λ level X	λF	λΕ	λD	λC	λΒ	λΑ
Formula	$\frac{\log \left[\frac{7}{4f}\right]}{\log \left(4f\right)}$	$\frac{\log \left[\frac{3}{2e}\right]}{\log (2e)}$	$\frac{\log\left[\frac{5}{4d}\right]}{\log\left[\frac{4d}{3}\right]}$	$\frac{\log\left[\frac{4c}{3}\right]}{\log\left[\frac{5}{4c}\right]}$	$\frac{\log(2b)}{\log\left[\frac{3}{2b}\right]}$	$\frac{\log(4a)}{\log\left[\frac{7}{4a}\right]}$
e.g. Tab. 1	$\lambda F = 0.984$	$\lambda E = 0.974$	$\lambda D = 0.973$	$\lambda C = 0.970$	$\lambda B = 0.967$	$\lambda A = 0.951$

Table 2. Formulae for calculation of the λ values (according to Edwards 1982) at all six levels using the numerical example in Table 1.

Based on the experience of Edwards it is advisable to variously weight the λ values of the different levels. We adopted his method and weighted the levels as follows: A, F fourfold and B, E twofold leaving C, D unweighted. From this the average λ values and standard deviations were calculated for each berry as well as for the entire sample on a given day. The latter defines a data point.

Limit regions The λ value of mistletoe berries varies around 1. Unripe berries tend to show values >1; ripe berries <1. The sketch in Fig. 2 shows the approximate limits within which the rhythmic shape changes in the berries take place. The following numerical values provide the basis of this construction: λ =1.05: f=0.646, e=0.855, d=0.962, T=1.000, c=0.974, b=0.878 and a=0.677; λ =0.95: f=0.678, e=0.878, d=0.975, T=1.000, c=0.962, b=0.854 and a=0.645.

Measurement precision Measurement of the photocopied berries requires a high level of precision. We obtained the best results when we pricked through the intersection point between the outline and the cross section line with a fine needle so that the berry diameter at the various levels could be measured on the reverse side of the paper without interfering shadows. In the first year (1990) 10 berries were measured per data point (day), but this was too few. In subsequent measurement periods we gradually increased the number of berries to double this. These two measures greatly increased measurement precision as the following values show. The average standard deviation of the mean for all harvest dates within the harvest period steadily decreased as follows: 1990, 8.2%; 1991, 4.9%; 1995, 3.8%; 1997, 2.5% and 1998, 2.1%.

In 1995 the standard deviation of the mean daily λ value had a maximum of 8% with about 90% of all values lying under 6%. In 1997 and 1998 all standard deviations of mean daily λ values were under 6%. The difference in these two years from that obtained previously was that a tolerance limit was set for the standard deviation of the mean λ value of a single berry: berries with standard deviations greater than 12% were regarded as deformed and the corresponding λ values excluded. In the years 1997 and 1998 this comprised 11% and 8% of all berries respectively.

In July 1995, berry harvests were carried out at various times of day. This showed that the scatter of the mean daily λ value was smallest between 8.00 and 9.00 am (see Table

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Jochen Bockemühl Bernerstr. 21 CH-4143 Dornach Switzerland unit, that we can comprehend as a turning outside in. What is developed by the growing plant from inside to outside and downwards from above, becomes lifeless and withers. But in the soil the process is spatially from the outside to the inside and from below upwards. The plant substance is comminuted and decomposed by small animals, fungi and bacteria and finally transformed into the universal humus that wholly integrates itself into the soil. This humus, which comes into being through the soil's life processes, is now outwardly open to the surroundings of the earth. It is a carrier of general effects of the elements via the simple minerals, water, carbon dioxide and warmth. This way it can be a fertile root environment from which the plant can take up what is suitable for it.

If comminution and degradation of plant residues proceed in water, no humus is formed. A uniform stimulating effect arises in water to which the growing plant is totally exposed, so that the uptake can hardly be species specific. But in the interplay with a healthy humus, plant roots can assimilate substances selectively.

When we follow the development of plants to their full expression and beyond to their differentiation into a multitude of substances during withering and decay and finally to the build-up of humus and new soil, we are following a central life process of the earth. We can experience this as a turning outside in; the outwardly grown plant becoming the integrated uniform soil formation process which offers the developmental conditions for new plant growth. We can consider these transformations as a respiratory process of the earth-plant world. In it we become aware in the realm of phenomena which, on another level, we can recognize as the life process that forms the basis for the development of our consciousness: in our waking consciousness we outwardly form from sensory perceptions our increasingly differentiated individual mental pictures. During sleep and in forgetting these pictures, the capacity to picture things turns inward in such a way that it unites with the context-forming universal power of the cosmos. In this way it can add to the perceptive capacity of thinking. To forget is an essential precondition for the ability to form ideas, which from this point of view is the same as the perceptive capacity. With it our potential for perceiving and picturing during daytime is extended.

Such a comparative consideration of the life processes of microorganisms and plants in the context of the earth is feasible only if we aim at an approach to nature that includes the necessary inner thinking complement. The point is to develop an inner perception of an experience that, together with the sensory perception, forms a reality in the individual's cognition.

We have to become conscious of an inner perception which is unconsciously present as an inner complement of a phenomenological approach to nature. This is comparable with the softly luminous yellow after-image that follows observation of deep blue. Only in such a dual way of looking at things does the aforementioned polar interrelationship between microbial processes and plant life become intelligible.

Our special thanks are due to Georg Maier who has through many years of cooperation so often given us confidence in such an approach to the world of appearances (Maier 2001). His approach, cultivated in his works in the field of physics (Maier 1986) and extended to aesthetics (Maier 1999), is to date an inspiring example for our own work in the fields of biology and ecology.

3). Therefore the period 8.00-9.00 am. can be regarded as optimal for berry harvest.

Time (h)	Number of days	Standard deviation (%)								
	of harvest (n)	Minimum	Average	Maximum						
08.00-09.00	7	2.4	3.8	5.6						
12.00-13.30	7	3.9	6.1	8.5						
16.15-17.30	7	1.4	6.2	12						
18.00-22.00	6	2.9	4.4	6.3						

Table 3. Standard deviation (%) of mean daily λ values as a function of four harvesting time windows

Other data sources Meteorological data were kindly supplied by the Basel-Binningen Meteorological Station, Venusstraße 7, CH-4102 Binningen, Switzerland. Relative sunspot numbers came from the Solar Influences Data Analysis Centre, Department of Solar Physics, Royal Observatory of Belgium, Avenue Circulaire 3, B-1180 Brussels, Belgium. Astronomical constellation data were obtained from Wolfgang Held, Mathematisch-Astronomische Sektion am Goetheanum, CH-4143 Dornach, Switzerland. These are the same data that form the basis of the ephemeris published by the Mathematisch-Astronomische Sektion am Goetheanum (Held 2001). Daily constellation data were calculated from the positions of all planets in the zodiac at 9.00 hours Central European Time.

Results of data analysis

Mistletoe berry shapes as path curves The mean daily standard deviation was maximally 6% with an exclusion of 8 or 11% of the berries in the years 1997 and 1998 (see above). Therefore the outline of mistletoe berries can generally be regarded as path curves. According to Edwards (1982) this cannot be taken for granted in the case of the seed-bearing parts of the plant.

1995 series of measurements In 1995, measurement of the shape parameter λ began on 5 July and ended on 2 December. Altogether, 48 λ values were obtained. Plotted as a function of time they showed a clear tendency to fall (Fig. 3) which reflects the gradual ripening process of the mistletoe berries. Linear regression analysis showed a significant correlation with time (p<0.001). By subtracting the regression equation, in order to increase the sensitivity of analysis to periodic processes, this drift was eliminated, thus giving rise to the trend-free shape parameter λ ' (Fig. 3). As the variability of λ ' is appreciably greater than the standard error of the measurements, one may suspect that further influences on λ ' would be identifiable.

For instance, correlations with meteorological parameters are conceivable. Therefore the λ ' values were compared with meteorological data from the nearest weather station (Basel-Binningen). The following parameters were considered: temperature (°C), pres-



Figure 3. Development of the shape parameter λ of mistletoe berries in the year 1995. Raw data (λ) and after subtraction of the first order trend ($\lambda' \pm$ standard errors).

sure (hPa), relative humidity (%), cloud cover (%), wind speed (m/s), each as 24 h mean values; precipitation (mm), hours of sunshine and global radiation (Wh/m²), each as absolute values. Any obvious annual trends in these data sets were likewise removed by linear regression. None of the meteorological parameters showed significant correlation (p<0.01, Spearman) with the λ ' values, not even with phase shifts of -5 to +5 days.

In connection with Edwards' work we had formulated the hypothesis that λ' values might correlate with moon-planet oppositions and conjunctions. However, as it was not clear *a priori* which planet to take into consideration and as additional phase shifts (Edwards 1992-1998) might arise, a statistical approach was adopted. To this end the constellation data for the 'planets' Mercury, Venus, Sun, Mars, Jupiter, Saturn, Uranus, Neptune and Pluto were parameterised as follows: for each of these heavenly bodies the date of moon conjunction and moon opposition as well as the day before and day after were assigned the numerical value 1. Quadrature dates with a day either side were numbered 3 and all other days were numbered 2. Then a Spearman rank correlation was performed on the parametrised constellation data and the λ' values, in the latter case allowing for a phase shift of up to 14 days. This yielded 135 correlation (9 planets multiplied by 15 λ' data sets). None of these correlations showed a statistical significance at the p=0.01 level.

Relative sunspot numbers can be regarded as a measure of cosmic ray activity. There is some evidence that not only geophysical but also biological and chemical processes may be influenced by solar radiation (for an overview see for instance Burns 1997). For this reason the λ ' values were also correlated with the relative sunspot number again with

These products were then represented in a sequence of their structural formulae. This can be exemplified by the reaction sequences of carbon dioxide assimilation into cell substance in green plants (spinach) and those of the degradation of carbohydrates to carbon dioxide during respiration of animal tissue (Schlegel 1992). Attention to the special qualities of the life processes has receded into the background, all the more so as the organisms had to be killed and studied analytically. In this way the objectification characteristic of thinking in physics became dominant in biochemistry.

A direction in which chemistry can be developed anew as a science of transformation processes in accordance with its true nature and aims was suggested by Steiner (1923). It is necessary to participate with mobile thinking activity in the sequence of appearances of a process and to develop an imaginative kind of cognition. This approach to the world involving inner activity may yield a new understanding of chemistry. In addition a special chemistry of life may be developed that comprises in equal measure both substance and form transformation processes (Bockemühl 2000). The present paper tries to approach the chemistry of life in the field of microbial life processes. It is the special feature of these life processes that they do not serve the growth and maintenance of a visible body as in higher organisms. Rather, they are active in shapeless transformations of substances which are invisibly enlivened through and through by microbes as process germs. The higher unit of these microbial life processes has quite another orientation: it is not an individual organism making sense in itself but is the biosphere of the earth as a whole in which it serves the other kingdoms of nature as environment forming forces.

8 Life processes in the plant-earth context

In conclusion, we would like to consider once again the relationship between plant development processes and microbial life processes. The character of these processes in the sense of interlinked supplementary life processes results from an imaginative consideration that acknowledges the mediating relationship between inside and outside in the context of the earth.

First of all we shall examine the rhythm of plant development. When a seed is ripe it is released from the developmental conditions of the mother plant. In the soil the seed germinates and in the course of the year develops outwardly into a plant that consists of highly differentiated substance. During its development the plant becomes an image not only of its inner nature (species) but also of the effects that are called forth by the environment. And the plant increasingly exhausts its formative potentialities. When flowering, the life process outwardly comes to an end and in this receding (comparable with respiration) the plant gains the capacity to form seeds and buds. The (spiritual) formative potentialities concentrate inwardly anew thereby largely loosening the seeds and buds from the connection with the earth. Here the plant process inwardly reaches its limits, but on the way it has gained the capacity to grow anew into particular forms that correspond to changed environmental conditions.

We now consider the development of the plant in its environment. With its green parts above the ground, the plant grows into the cosmic periphery towards the light of the sun. Polar to this the roots grow down into the soil. In their polarity both regions form a higher and/or relationship groups. Therefore, cell morphology is of little significance for classification. However, the special metabolic capacities (kinds of substrates and metabolic products) of the cells are species and genus-specific. These can only be determined experimentally by the use of pure cultures. On the basis of results obtained in this way we understand the species as a specific 'process-form' which is active in a chemical environment that corresponds to its specific metabolic capacities. And we consider the shapes of the cells as 'form of life-types' which have a functional relationship to the physical properties of their habitats (Pfennig 1995). With this we now compare the characteristics of the eukaryotic unicellular organisms (e.g. amoebae, ciliates, flagellates, diatoms; Fig. 2b). The shapes of their cells are so rich and specifically differentiated, that each species can be recognized and determined purely by morphology. In contrast to this, the metabolic capacities are comparatively uniform and serve primarily the growth of the cells. The eukaryotic cell with its polarity of nucleus and cytoplasm lives comparatively more independently of its environment and has developed the capacity for the action of species-specific formative forces at the cellular level. Only rarely, as in the case of the cellular slime moulds among the Myxomycetes, does a multicellular form of higher order occur. As the division of the cells usually leads directly to multiplication, the formation of a multicellular organism becomes impossible.

The relationship between specific cell shape and metabolic activity in eukaryotes compared with bacteria has to be considered as polar. The eukaryotic cell appears microscopically as a highly differentiated formation; its metabolic process is internal and remains invisible. In the case of bacterial processes we perceive the decomposition of the substrates (e.g. soil, sediment, waste water); the bacteria as process germs remain indiscernible. Their microscopically visible cell shapes are, as discussed above, not comparable with the cell shapes of the eukaryotes. We can fully understand the bacterial process germs only if we correlate their appearance with our inner perception (intuitive faculty) that we formed from the sequence of appearances of the substrate transformation process to which they belong.

7 Chemistry and microbial life processes

Chemistry by its very nature is a science of the transformations of substances. These are chemical movements in the sense of changes and they mostly depend on a liquid medium. As we have shown, it requires an approach different from that of physics. Only in this way does it become clear how, chemically speaking, a substance points beyond itself. This applies whether we consider it as the product of a synthesis or as the starting material for new substances (through analysis or synthesis). It follows that a chemical substance is connected with other substances not only temporally but also through its surroundings. Therefore, it is not necessary to restrict chemistry to the study of the synthesis and analysis of single substances. We may also consider transformation processes that, viewed analytically, comprise whole reaction sequences as well as belonging to the context of life. In the past century biochemistry was developed from a quite different point of view. With more and more refined analytical methods, certain life processes of organisms became separated into single reactions of consecutive intermediary and final products.

provision for a ± 14 day phase shift. None of these correlations yielded statistical significance at the p=0.01 level.

In connection with these 'negative' results the question arises as to whether there could be other periodic processes present. There are too few data points for a Fourier analysis and autocorrelation is problematic because of the unequal periods of time between the individual sample points. For these reasons a different approach was chosen. Thirty one sinusoidal curves with wavelengths between 3 and 33 days were constructed and once again a Spearman rank correlation coefficient was calculated between these sinusoidal curves and the λ ' values (on the original time scale as well as with a phase shift of up to 30 days). Significant correlations (p<0.01) were obtained only with a wavelength of 10 days (p=0.0009 for a phase shift of 4 days, p=0.0034 for a phase shift of 3 days).

This periodicity can be related to the lunar-zodiacal-trigonal rhythm. The moon's sidereal cycle is 27.32 days, i.e. the moon stays in a zodiacal constellation or sign on average 2.28 days. Every 9.11 days the moon enters a constellation or sign which belongs to the same 'element' (see table 4). This relationship of the zodiacal constellations or signs to particular 'elements' handed down by tradition has acquired practical importance in agriculture in the work of Thun and Heinze (1973).

Element:	Fire	Air	Earth	Water
Constellation	Aries	Taurus	Gemini	Cancer
or sign in	Leo	Virgo	Libra	Scorpio
zodiac	Sagittarius	Capricorn	Aquarius	Pisces

Table 4. Relationships of zodiacal constellations and signs to the 'elements'

When the λ' values are given as a function of the moon position in front of the corresponding groups of zodiacal constellations or signs one might suspect that there is a relationship between them (Fig. 4). Whereas when the moon is in front of constellations of the elements fire and earth the tendency is to low λ' values, they tend to be high when it is in front of air and water element constellations. As a function of the zodiacal signs there is a corresponding shift (Fig. 4). However, these differences cannot be verified statistically.

These initial results nevertheless interested us sufficiently to test in two subsequent years (1997 & 1998) the hypothesis that there is a relationship between the λ' values of mistletoe berries and the moon's position in the zodiac, grouped according to elements. The period of data acquisition was limited to between mid-October and mid-December partly because of available resources and partly because of its relevance to actual practice as the mistletoe harvest for the preparation of *Iscador* at the Hiscia Institute takes place in November.



Figure 4. λ ' values (mean and simple and doubled standard errors) of mistletoe berries (1995 harvest) as a function of the position of the moon in the zodiac, grouped according to the elements of a) zodiacal constellations and b) zodiacal signs.

1997 and 1998 series of measurements The measurement periods for 1997 (18 λ values) and 1998 (22 λ values) were 17 October to 8 December and 12 October to 15 December respectively. There was no significant linear trend in the course of time in the raw data (Figs. 5 and 7). This accords with the 1995 data as the 1997/1998 measurement periods correspond to the last part of the plot of 1995 data (approx. days 100-160 in Fig. 3) which has a more or less horizontal linear regression line. However, for the sake of comparability, the 1997 and 1998 data sets were also standardised by subtraction of the linear regression equation from the raw data. This yielded the λ ' values for the corresponding periods of time (Figs. 5 & 7).

As with 1995 data, the Spearman correlations of the λ ' values with the trend-free weather data (see above) were calculated with phase shifts of up to ±5 days. Again neither in 1997 nor 1998 were there significant correlations (p<0.01, Spearman). This was also true of the correlation of the λ ' values with the relative sunspot counts (likewise with phase shifts of up to ±14 days).

The periodicity of the λ' values, which is immediately noticeable from inspection of Figs. 5 and 7, was obtained using the method of correlation with sinusoidal curves described above. Significant correlations (p<0.01) occurred with curve periodicities of 9 and 10 days in 1997 but only 9 days in 1998.

Our hypothesis concerning a coincidence of λ ' values and moon positions in the zodiac can be confirmed (Figs. 6 & 8). Whereas when the moon is in front of constellations of the elements fire and earth the tendency is to low λ ' values, they tend to be high when it is in front of air and water element constellations. And again, as a function of the zodiacal sign there is a corresponding shift (Figs. 6b & 8b).

We realize that the cellular organization of a living organism is a pictorial expression of its actual and possible functions and is therefore of fundamental importance. This is also apparent in the case of the following characteristics. In bacteria only a few types of cell shape exist (Fig. 2a: sphere, rod, spiral, filament) that occur in almost all physiological





The microscopic and macroscopic images of the process as well as the experiences of growing and decaying that are connected with them are in a complementary relationship to each other. If we bear in mind that we can properly think about the microscopic cellular level only with respect to the connected level of macroscopic substrate degradation, then the macroscopic level deserves our special attention.

6.2 Effect of culture conditions

We took a static bacterial culture of constant volume as an example of a microbial transformation process. The developmental stages of growing, resting and decay are exclusively caused by the continuously changing conditions in the culture medium: the growth substrate is increasingly used up by the multiplying bacteria and metabolic products are accumulated accordingly. Therefore the developmental stages of the culture are a consequence of the metabolic activity of the bacteria and not an expression of a bacteriaimmanent 'time dependent form' as exists in higher plants. Only if we consider the entire context of life on earth does a 'time dependent form' emerge also for the microbial life processes (see concluding section).

If we study the bacterial degradation process in an open flow system of continuous culture (Pfennig and Jannasch 1962) we are able to obtain a steady state. In this system, variations take place only if the culture conditions are changed. In the steady state the culture is time-invariable, although the process of course proceeds in time. In this case the microbial process corresponds to the chemical transformation process of a candle flame. The latter can be considered as a 'flow-form' that is seen as a time-invariable flame (Bockemühl 1967).

6.3 Prokaryotic and eukaryotic unicellular organisms

Previous papers (Pfennig 1984b, 1995) described in detail how two basic types of micromorphological cellular organization exist in living organisms: the prokaryotic and eukaryotic cell types. The prokaryotic cell organization is an essential feature of all bacteria, while all other uni- and multicellular living beings have a eukaryotic cell organization. The prokaryotic cell is not differentiated into a membrane bound nucleus and cytoplasm; its simple shape is maintained by turgor pressure against a relatively stiff cell wall. As a process germ-cell the prokaryotic cell is the center of the unlimited open surroundings in which it is effective.

In contrast, the eukaryotic cell is polarly organized: it has a membrane bound nucleus and a surrounding cytoplasm with organelles. The cytoplasm also contains a cytoskeleton which has several functions: it maintains the shape of the cell and serves for the movement of internal cell structures, e.g. vacuoles, but also the movement of the cell itself. Many groups of eukaryotes ingest food particles by endocytosis or phagocytosis. The ability for endocytosis allowed the eukaryotic cell to take up prokaryotes with certain metabolic capacities from the environment. Membrane bound endosymbiotic cytoplasmic organelles like mitochondria and chloroplasts are understood to have been formed in this way. Significant metabolic activities present as prokaryotic cells in the environment thus became individual capacities in the cytoplasm of eukaryotic cells.







Figure 6. λ ' values (mean and simple and doubled standard errors) of mistletoe berries (1997 harvest) as a function of the position of the moon in the zodiac, grouped according to the elements of a) zodiacal constellations and b) zodiacal signs.



Figure 7. Development of the shape parameter λ of mistletoe berries in the year 1998. Raw data (λ) and after subtraction of the first order trend ($\lambda' \pm$ standard errors).



Figure 8. λ ' values (mean and simple and doubled standard errors) of mistletoe berries (1998 harvest) as a function of the position of the moon in the zodiac, grouped according to the elements of a) zodiacal constellations and b) zodiacal signs.



Figure 1. Rhythm of growth and division in bacteria.
1 and 4: Cell that originated by division grows to twice its length.
2 and 5: Grown cell divides in the middle and daughter cells become separated: 3 and 6

multiplication process as a continuous dissolution of form. The cells and their metabolic activity gradually spread throughout their substrate. The concomitant rhythm of substrate uptake and metabolic product release remains microscopically invisible, but the surroundings of the cells are continuously altered chemically.

The visible changes of the substrate when we follow them experimentally, for instance in the liquid medium of a large bacterial culture, are important *macroscopically*. The cells are invisible but cause a turbidity of the medium. The degradation of the growth substrate is often accompanied by specific color changes, unpleasant smells and changes in the consistency of the whole culture medium, e.g. through slime formation.

We now have to correlate these changes which we experience as decay with the microscopic pictures of the cells. In most cases we find that during the period of strongest substrate degradation the bacteria undergo their fastest multiplication. They look well formed and are experienced as being healthy.

Only at the end of the degradation process when the culture has reached the specific state towards which the whole process was heading do the bacterial cells begin to lose their characteristic form. They become irregular in shape, stop growing and multiplying and finally disintegrate.

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It is apparent from the preceding paragraph that chemical reactions as temporal transformations demand a 'fluid' approach, although they are always realized only by mental experiences in the physical world.

If we intend to understand the specific qualities and the directed activity of an ongoing microbial process as an expression of its essence, we have to approach it according to the means of the element of warmth. The essence discloses itself to our self-experience only if we convert the multitude of the sense appearances of the entire process into an inner perception (intuitive faculty). We have intentionally to identify ourselves with the process, so that we are able to reproduce it again with this new faculty (multiplicity in unity).

Microbial transformations in the laboratory are usually established to achieve a certain result. From one's insight into the entire process it is necessary therefore to arrange the starting conditions in such a way that this result is achieved. Therefore we may say that the final effect of the process is caused by the arrangement of the initial conditions for a given culture. There is one course of time proceeding from the past to the future that can be accompanied by sensory perception. Opposed to this is a second one acting from the future to the present. The latter is accessible only to inner cognition. This course of time is complementary to the growth process of a plant in such a way that the plant continuously remains a perceptible whole. The inner driving force of the formative principle of the plant comes together rhythmically with the gradually emerging outward shape. Only if we become one with the entire process in our inner perception (intuitive faculty) are we able to experience the specific arrangement of its essence outside of the course of time in the state of no change (Bockemühl 1967).

6 Transformation processes and cellular organization

6.1 Microscopic and macroscopic levels

From the phenomenological point of view we have to consider two levels of perception in microbiology: the microscopic and the macroscopic. There are transformation processes at both levels. When using the microscope we can see the microorganisms multiplying but on the macroscopic level we generally only observe the transformation of organically formed substances in soil, water or sediment.

Let us first consider bacterial cells multiplying on a semi-solid culture medium under the microscope (Fig. 1). A rod-shaped cell that has just originated by cell division grows to twice its length while the cell width remains unchanged. Then the cell divides in the middle and the two daughter-cells again grow to twice their lengths, divide again and so on. We perceive a rhythmic change of cell growth and cell division that results in the multiplication of the cells in geometric progression until the growth substrate is used up. We obtain a sequence of images that we connect to the process of multiplication with our mobile pictorial thinking activity. Within this rhythmic process each division represents the end of the previous growth process and the beginning of the next; the ending of one step allows the beginning of the next.

In this way *microscopic* study reveals only the simplest rhythm of life. We become aware of a process of cellular multiplication that does not go beyond the individual cells – i.e. no higher multi-cellular organism is being formed. Therefore we experience the

A non parametric analysis of variance (Kruskal-Wallis) gave globally significant statistical differences (p=0.0038 (1997) or p=0.0014 (1998) for the relationship to the zodiacal constellations and p=0.0037 (1997) or p=0.0014 (1998) for the zodiacal signs).

Combination of the series of measurements in 1995, 1997 and 1998 A detailed statistical analysis was carried out on the λ ' values from the years 1995, 1997 and 1998 combined (n=88). The pooled data were shown statistically not to depart from a normal distribution (p=0.51, χ^2 test) thus allowing parametric statistical methods to be used. Analysis of variance using the F-test showed globally significant differences (p<0.0000001, for both zodiacal constellation and zodiacal sign groupings, Fig. 9).



1995, 1997 & 1998 harvests

λ'

Figure 9. λ ' values (mean and simple and doubled standard errors) of mistletoe berries (1995, 1997 and 1998 harvests) as a function of the position of the moon in the zodiac, grouped according to the elements a) zodiacal constellations and b) zodiacal signs.

Comparison of pairs (LSD-test, least significant difference) amongst the four elements gave no significant differences (p>0.1) in the case of the zodiacal signs (Fig. 9b) between either the two maxima (fire and water) or the two minima (air and earth). However, comparing all maxima and minima with each other, i.e. fire or water with earth or air, gave significant differences in all cases (p<0.002).

The same applies to the grouping according to zodiacal constellations (Fig. 9a). Individual comparisons amongst the four elements yield no significant differences (p>0.1) between the two maxima (air and water) or the two minima (fire and earth). However, comparing all maxima and minima with each other, i.e. fire or earth with water or air, gave significant differences in all cases (p<0.002). This in fact means that the λ ' values of mistletoe berries when the position of the moon is in front of the zodiacal constellations for the elements fire and earth are about 4% lower when compared with the corresponding values for the elements air and water. The variations are thus quantitatively modest

but fall within the limits of detection (compare Fig. 2).

Taking λ ' values as a function of the individual zodiacal constellations, one obtains the expected saw-tooth pattern (Fig. 10). Thus there appears to be no difference between the individual members of a group of elements (Table 4). Qualitatively the same result is obtained when grouping the data according to zodiacal signs.



λ' 1995, 1997 & 1998 harvests

Figure 10. λ ' values (mean and simple and doubled standard errors) of mistletoe berries (1995, 1997 and 1998 harvests) as a function of the position of the moon in front of the zodiacal constellations.

Discussion

In connection with the findings of Edwards (1982) the investigation of shape changes in ripening mistletoe berries started with the hypothesis that moon-planet rhythms would be detectable. This hypothesis could not be verified in any of the years of this investigation. However, the hypothesis proposed in 1995 that the shape changes can be correlated with the position of the moon in the zodiac was confirmed in the subsequent years of investigation, 1997 and 1998.

As neither harvest nor measurement were as a rule carried out blind, the question arises as to what extent these results could unconsciously be influenced by the experimenter. Such a 'bias' can be excluded with regard to the year 1998 as the berry harvest in that year was carried out by someone who was not informed about the working hypothesis. As the results for 1997 and 1998 are essentially the same, an unconscious influence at berry harvest, for instance in the sense of an unconscious pre-selection of berries that 'fit', can be ruled out with high certainty.

5 Approaches based on the elements

Chemical reactions of substances are often accompanied by changes of aggregation state, i.e. with the transitions from solid to liquid (dissolution) or from liquid to gaseous state. In all cases these are also associated with an increase or decrease in warmth. Approaches to chemistry and microbiology that are complementary to the usual way of thinking in particles (molecules) start with the insight into the qualities of the states of substances which are usually called aggregation states. This designation implies different kinds of aggregation of the molecules in each element. At best this is an appropriate designation for the solid element. The conception tries to explain the states of the other elements by the kind of cohesiveness of their molecules. However, we can do justice to these different states only if we can find in our own relation to them the appropriate approach to their different qualities.

We comprehend the earthly solid element with our fixed mental pictures, with our ability consciously to grasp something in order to separate it from its surroundings and place it before us. Such pictures are thereby accompanied by inner experiences of our physical body. We become self-confident through them, i.e. we develop a 'self-experience in the physical body' (Steiner 1923). This is the basis for our usual representational mode of cognition. In chemistry this happens when we explain processes by the structural formulae of successive individual substances. However, we are also dealing with transformations to which we cannot do justice through a representational mode of cognition. We need a fluid approach.

For the approach to the fluid element we have to accord with the manner in which water appears, i.e. in a great many changing pictures which correlate with the surroundings. We have to follow the transitions from one pictorial context to the next in a perceptive way of thinking. The same is necessary for all chemical transformations that proceed in the liquid phase.

We experience a corresponding capacity in our instinctive consciousness of the vital functions of our body (Steiner 1910), e.g. in the way that after-images of outer perceptions arise like answers from our inner being. In addition, we find the capacity in our self-confidence that relies on our own thinking activity while following and experiencing a sequence of appearances. This way we aim at an imaginative form of cognition.

Many microbial processes are accompanied by the generation of smells and gases. If we become aware that we are not being confronted by smells and gases but rather steeped in their medium and perceiving them directly with our breathing, we are then approaching them in a way that accords with the element of air. Our entire mood is changed by the perception of a smell and we experience the inner quality of a substance that has become volatile. Certain instinctive reactions may take place and the life processes of our body are also affected. For instance the pleasant smell of a delicious meal makes our mouth water. On the other hand evil smells may turn our stomach so that we lose our appetite. Smell and color perceptions are experienced in a similar manner. Therefore, we turn our attention to color changes of microbial processes with the approach that is relevant to the element of air. This requires an unselfish devotion to the inner experience of an outer appearance as in the case of observing a gesture.

	in light	in light	freshwater	or seawater	neutralized	5	plant parts	broken down	to fibers	green to	black		red-brown	deposit on	walls in light		hydrogen	sulfide	
ut air	ation impossible		sea water		neutralized	4	plant parts	broken down	to fibers	green to	black		no gas	production			very strongly	of hydrogen	sulfide
witho	complete degrad	in darkness	fresh water		neutralized	ŝ	plant parts	broken down	to fibers	green to	gray-brown	to black	vigorous gas	production	(CH_4, CO_2)		musty,	faintly of	hydrogen sulfide
					acidifying	7	plant parts	little altered,	limp	green to olive	green		acid self-	preserved			like silage		
1 air	idation possible	in darkness			neutralized	1b	forms of	plant parts	dissolve	greenish to	light ochre,	turbid					musty earthy		
with	complete degra	in light			neutralized	1a	forms of	plant parts	dissolve	from green to	light ochre,	turbid	green	coloration at	end,	microalgae	musty earthy		
		Initial	Conditions			Type	plant	substance		color change			other changes				smell		

Table 1. Types of degradation process in water under various initial conditions

The hypothesis that there was an unconscious influence during determination (measurement) of the λ values cannot at present be ruled out with absolute certainty. However, we consider the probability of this to be small as the data for the 1995 investigation, with its hypothesis at that time of a fortnightly rhythm, shows not trace of such a periodicity. Greater certainty on this issue could be obtained for instance with a 'blind' determination of the λ values or through non-blind measurements with other expectations relating to possible results.

Naturally other questions arise too: for example one might be whether the phenomena shown here can also be observed with mistletoe berries of another tree. In this investigation the same tree was used in all three years for reasons of comparability.

A further question would be whether it could be determined empirically if the mistletoe berry shape is correlated more with the zodiacal constellations than the signs or vice versa. This could be achieved by a precise 'survey' of that part of the zodiac where the boundaries of neighbouring zodiacal constellations and signs lie the greatest distance apart. For instance, a region of interest would be the sector $140^{\circ}-150^{\circ}$ where according to the correlation with the zodiacal constellations low λ values would be expected (constellation Leo, fire element, Fig. 9a) but where according to the correlation with the zodiacal signs high λ values would be expected (zodiacal sign Leo, fire element, Fig. 9b). Unfortunately in the region referred to there was not a single measurement.

All such questions will be addressed in a subsequent paper. According to current understanding the results presented nevertheless point to the possibility of a correlation between moon-zodiacal constellations and biological phenomena.

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Translated from the German original by David Heaf

Bovine spongiform encephalopathy and uric acid

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This is a report of my recent work at the University of Bath, U.K (please see Science Group of the Anthroposophical Society of Great Britain Newsletter, March 2002). The results presented are due to be published in a more complete form later in 2003. Because of the direct connection of this work to an insight put forward by Rudolf Steiner, I have decided to write a preliminary report from an anthroposophical perspective.

Rudolf Steiner and 'mad oxen'

Rudolf Steiner predicted the possibility of a disease similar to bovine spongiform encephalopathy (BSE or 'mad cow' disease) in one of his lectures (GA348). The relevant paragraph, as it appears in English translation, is quoted below (Steiner, R. 'Health and Illness', Anthroposophic Press, 1983 pp. 82-85).

'Consider a cow or an ox. After some years the flesh within it has been entirely replaced. With oxen the exchange takes place even faster than with human beings. A new flesh is therefore made. From what did this flesh originate, however? You must ask yourself this. The ox itself has produced the flesh of its body from plant substances. This is the most important point to consider. This animal's body is therefore capable of producing meat from plants. Now, you can cook cabbage as long as you like, but you won't turn it into meat! You do not produce meat in your frying pan or your stew pot, and nobody ever baked a cake that became meat. This cannot be done with outer skills, but taken fundamentally, the animal's body can accomplish inwardly what one can't do outwardly. Flesh is produced in the animal's body, and to do this forces must first be present in the body. With all our technological forces, we have none by which we can simply produce meat from plants. We don't have that, but in our bodies and in animal bodies there are forces that can make meat substance from plant substance.... Now imagine that an ox suddenly decided that it was too tiresome to graze and nibble plants, that it would let another animal eat them and do the work for it, and then it would eat the animal. In other words, the ox would begin to eat meat, though it could produce the meat by itself. It has the inner forces to do so. What would happen if the ox were to eat meat directly instead of plants? It would leave all the forces unused that can produce the flesh in him. Think of the tremendous amount of energy that is lost when the machines in a factory in which something or other is manufactured are all turned on without producing anything. There is a tremendous loss of energy. But the unused energy in the ox's body cannot simply be lost, so the ox is finally filled with it, and this pentup force does something in him other than produce flesh from plant substances. It does something else in him. After all, the energy remains; it is present in the animal, and so it produces waste products. Instead of flesh, harmful substances are produced. Therefore, if an ox were suddenly turn into a meat eater, it would fill itself with all kinds of harmful substances such as uric acid and urates. Now urates have their specific effects. The specific effects of urates are expressed in a particular affinity for the nervous system and the brain. The result is that if an ox were to consume meat directly, large amounts of urates would be secreted; they would enter the brain, and the ox would go crazy. If an experiment could be made in which a herd of oxen were suddenly fed with pigeons, it would produce a completely mad herd of oxen. That is what

We experience the blackening of the degradation media as a gradual cessation of the transformation activity. The process activity ceases and loses further direction which is comparable with what we experience on a night without the light of the moon or the stars. Provided the darkness does not stop us completely, we inwardly seize new opportunities to proceed. In a comparable way, the black turbidity may become the starting point for microbial transformation processes in a new direction if correspondingly modified environmental conditions become established.

Sequence of smells: At first the smell in all three types (3 to 5) is caused by the inoculation material: a slightly moldy smell. This slowly intensifies towards the moldy-musty; we experience this as stifling and obstructive to breathing. Together with the conversion of color to black we notice an increasing smell of hydrogen sulfide. It is a repulsive musty-sweetish smell that we can tolerate only from some distance. This smell provokes a strong feeling of nausea in the epigastric region and impairs our feeling for life. We have to turn away from this oppressive stifling-sweet atmosphere. We can clearly differentiate the hydrogen sulfide smell from both the disgusting carrion or putrefaction smell and the slightly musty-aromatic smell of rotting fruit and vegetables.

Degradation experiment of type 5 with illumination: The degradation processes of types 4 and 5 occur in the lower oxygen-free regions of marine aquatic environments, below the aerated and illuminated layers of water. Therefore these processes mostly remain unobserved. However, there are localities where the degradation of plant residues linked to dissimilatory sulfate reduction reaches up to water layers with access to sunlight. In our degradation experiment the corresponding places are the side walls of our glass bottles. Here illumination gives rise to transformation processes that become apparent by their rose red, ruby red or brown red colors. The anoxic degradation processes ending in black colors reveal a latent receptivity to light and proceed to red colors visible from some distance. In these colors we experience the spiritualizing activity of the sulfur process and its terminal light-dependent part. The different types of microbial degradation processes of plant residues in aquatic environments are briefly summarized in Table 1.

4.3 Microbial transformation of animal body substances

In the presence of air, substances of the animal world are degraded relatively quickly and completely. Inorganic nitrogen compounds are formed in corresponding quantity (types 1a, 1b). In the absence of air, only degradation processes of the types 3 to 5 are possible as fermentation acids (type 2) become neutralized by ammonium ions simultaneously liberated (the exception being milk because of its lactose content).

In addition to the phenomena described for the degradation of plant residues, additional insufferably evil smells arise in the case of animal substances. They indicate the presence of organic nitrogen-sulfur compounds (mercaptans) as well as nitrogen compounds known as ptomaines (agmatine, cadaverine, putrescine, skatole and others). In addition, very toxic compounds may appear (e.g. botulinum toxin) which are formed by the anaerobic bacteria involved in protein degradation. On account of these changes it is understand-able why anoxic degradation of animal substances is avoided under all circumstances.

formed into indistinct pictures of amorphous matter. Finally we free ourselves completely from such pictures and follow in thought connections the appearances of the turbid liquid medium. The direction of transformation is given by the arrangement of the experimental conditions: it is the gradual decrease of the turbidity by respiratory degradation giving rise to gaseous products, primarily carbon dioxide.

Transformation of colors: At first the fresh leaves are vivid green. As they become limp the green turns into dark green and later olive green. As the light grayish turbidity of the medium increases, the greenish colors become greenish ochre and finally light ochre.

We experience with the succession of these colors the decomposition of the plant leaves which reminds us of the discoloration of vegetation in the fall. With the conversion of olive green to brightening ochre we free ourselves from the world of facts and experience a slight expansion into the width of the universe. The brown colors typical in withering leaves in nature do not appear because in an aqueous medium the whole process proceeds relatively quickly.

Sequence of smells: The well aerated degradation experiment smells only slightly. The initial moldy smell comes from the inoculation material and gradually decreases. Towards the end of the process a faint earthy smell develops and is experienced as 'aged spiciness'.

Through a multitude of specific processes the entire degradation process reaches an indifferent resting stage with the capacity to become the basis for new developments.

4.2.2 Degradation processes in the absence of air (types 2-5)

Arrangement of process conditions: If we exclude the access of air to a degradation process, we experience this as an isolation from our context of life. The transformation processes are unable to respire from the atmosphere and therefore turn to respiration reactions within the medium in which they are contained. All processes must gradually cease, which we experience as suffocation. Inside the bottle an atmosphere becomes established that excludes all higher forms of life. In the case of the degradation experiment of type 2, the plant material quickly undergoes an acid fermentation. Although the plant leaves become limp, they retain their form and are acid-preserved. Thus a spiritualization of the residues is prevented. This is different in the case of the degradation experiments of types 3 to 5 that are maintained in the neutral pH range. The conversions of forms and colors take a similar course in each case and result in a spiritualization of the plant material. As long as air remains excluded the hostile atmosphere of these processes cannot be overcome.

Transformation of forms: At first the changes of the fresh leaf forms proceed similarly to those in the presence of air: the leaf parts become limp and fall apart into smaller and smaller pieces. Forms disappear and only amorphous residues and the finest fibers remain. No further disintegration occurs so that in the end a muddy sediment is formed.

Transformation of colors: The vivid green of the fresh leaves changes to dark green and olive. The light grayish turbidity caused by the inoculum gradually increases and becomes dark gray. Later this turns into a uniform blackish turbidity within which remnants of leaf substance are noticeable.

would happen. In spite of the gentleness of the pigeons, the oxen would go mad.'

Project rationale

The extract above has stimulated discussions among anthroposophists about the possible role of uric acid in bovine spongiform encephalopathy (BSE) or 'mad cow disease'. A number of people suggested that there was a significant similarity between Rudolf Steiner's description of the appearance of madness in bovines when they are fed meat instead of grass and the way by which BSE is supposed to have spread (details below). As no one had tackled this problem experimentally, I decided to create a project to answer the following fundamental questions: (1) Is the disease described by Steiner the same as BSE? (2) Does uric acid accumulate in the brains of cattle with BSE (3) Can uric acid cause neurodegeneration as seen in BSE.

BSE background

BSE is a prion disease In 1923 when Steiner gave his lecture, scrapie, a neurological condition of sheep had already been known for some time (the first recorded case of scrapie in the U.K. was in Bath, as reported in the Agricultural Improvements Society of Bath, 1788). A similar condition in cattle, however, either did not exist or was so rare that it was not recorded. It was only in the last 20 years that we have seen the rise of a scrapie-like neurological condition in cattle. The suspected reasons for the emergence of the new cattle disease have intriguing parallels with the scenario painted by Steiner. For many years prior to the mid 1970s, British cows were fed a protein supplement made out of meat and bone-meal derived from ruminant animals (Ford, 1996; Hahn, 1999). In the late 1980s, 'mad cow disease' reached epidemic proportions in the British Isles. Many people made the connection between the feeding of meat and bone meal to cows and the rise of the disease but the proposed reasons for the spread of this disease had nothing to do with the feeding of the meat and bone meal supplement *per se*. Instead, the supplement appeared to be acting as a medium through which a particular kind of infection could be passed on.

Today, many scientists argue that BSE and diseases like it are caused by an aberrant cellular protein PrP^{c} which, through a change in its three dimensional structure loses its normal function, accumulates and becomes lethal $PrP^{s_{c}}$ (Prusiner, 1989; Meyer et al., 1986; Oesch et al., 1985; Pan et al., 1993). These diseases are collectively known as 'prion' diseases. The word 'prion' was introduced to describe the novel proteinaceous, infectious agent believed to cause diseases like scrapie in sheep, BSE in cows and CJD in man (see Prusiner, 1982). It appears that the abnormal prion protein ($PrP^{s_{c}}$) can itself catalyse the structural change in PrP^{c} . If it enters the nervous system, it causes the normal PrP^{c} present in nervous tissue to change its structure to $PrP^{s_{c}}$ and become lethal. This is the accepted basis of prion infection. The abnormal protein can also be generated 'spontaneously' (i.e. by unknown causes), by an inherited mutation or by transmission.

In the early 1980s, the usual method of pre-treating meat and bone-meal supplement by high temperatures and organic solvents, was abandoned for economic reasons. This is thought to have encouraged the survival of the infectious prion agent within the feed. The

source of the prion agent in the feed is not known. One theory proposes that the prion protein mutated spontaneously in an individual cow, that cow was slaughtered and her remains contaminated with PrP^{sc} were included in the feed. This was then eaten by other cows and caused the disease BSE.

Prions will not cause disease on their own In spite of the currently held belief that prion diseases are caused by PrP^{sc}, there is some uncertainty as to whether the accumulation of the PrP^{Sc} per se can account for the symptoms. Experiments in living animals (in vivo) and cell culture systems (in vitro) have suggested that the presence of PrPSc alone is not sufficient to cause the disease. Mice have provided good in vivo experimental models and much of our knowledge of prion diseases is derived from experiments with these animals. Genetically engineered mice lacking the normal PrP^C protein (PrP-knockout mice) were created (Büeler et al., 1993) and since these mice lacked PrP^c, they were unable to perform the conversion of their own PrP^C to PrP^{Sc}. Such mice were resistant to prion disease and showed no neurodegeneration on infection. An ingenious transplantation experiment carried out in Zürich showed that PrP^C expression was absolutely necessary for prion disease-type neurodegeneration to take place (Brandner et al., 1996). Transplantation of embryonic tissue from a genetically engineered mouse expressing high levels of PrP^c was made into the PrP-knockout mouse brain. The transplanted tissue was then infected with mouse scrapie. This transplanted tissue generated PrP^{sc} and showed signs of neurodegeneration while the surrounding brain tissue (lacking PrP^c) remained whole. PrP^{sc} from the transplanted tissue apparently spread throughout the brains but did not cause neurodegeneration in any regions where PrP^C was not present. Thus it is clear that neurones must express PrP^C in order to be killed by PrP^{Sc}. Prion infections are therefore not a straight case of 'poisoning' by a toxic substance. The disease symptoms depend on a process, or a number of processes, taking place in a co-ordinated way.

Neurodegenerative processes in prion disease What are these other processes? As stated above, the rapid neurodegeneration in prion diseases follows from an increased deposition of abnormal PrP^{sc}. PrP^{sc} stimulates the conversion of PrP^c to more PrP^{sc}. After conversion, PrP^C is no longer able to perform its normal cellular function, which is to act as an antioxidant enzyme (Brown et al., 1999; Wong et al., 2001a; Wong et al., 2001b; Sassoon, 2002). There is now a considerable amount of evidence to show that cells in which PrP^{C} has been inactivated are less able to respond to the presence of highly reactive molecules called free radicals (Hogg, 1998). Free radicals react very rapidly with other molecules and can damage proteins, DNA and lipids. They are often chemical derivatives of oxygen and are constantly being produced in the living brain. Some free radicals arise as useless biproducts of normal biochemical processes while others are generated for useful purposes, for example nitric oxide (NO) for neurotransmission. The superoxide (O_{2}^{-}) radical is generated as part of the inflammatory response and immune defense in the brain. Excess free radicals are inactivated by molecules called antioxidants, which convert them into harmless chemicals like oxygen and water. PrP^c is one such antioxidant. The presence of too many free radicals creates a condition known as oxidative stress and can result in cellular damage. Thus with the conversion of PrP^c to PrP^{sc}, brain cells become less well protected against oxidative stress.

posal of the digested sludge causes problems in many communities. In nature we find the degradation processes leading to methane formation particularly in the muddy sediment of eutrophic ponds and pools with luxurious plant development. These processes become apparent by the bubbles of marsh gas that frequently rise to the surface.

Type 4 and 5: In marked contrast to fresh water, sea water contains very high sulfate concentrations $(2.6 \text{ g } 1^{-1})$. In the marine environment, the anoxic decomposition of plant residues is preferentially linked, therefore, to dissimilatory sulfate reduction leading to hydrogen sulfide as the terminal product. Even in bacteria tolerance of this poisonous metabolic product is limited with the result that the whole process gradually inhibits itself. In nature we find these degradation processes particularly in lagoons that are rich in algae at shallow sea coasts or in oceans as well as under the surface of sandy shallows and adjacent salt marshes.

Type 5: If the hydrogen sulfide forming degradation processes proceed upwards to anoxic water or sediment layers receiving sunlight, particular bacterial transformations become possible. They attract our attention by their conspicuous rose red, purple red, or brown red coloring of water and sediment. In these so-called sulfureta, red sulfur bacteria perform an oxidation of hydrogen sulfide to sulfur and sulfate independent of oxygen and dependent on light (Pfennig 1989). To a much lesser extent similar processes occur in the summer and fall in muddy freshwater ditches, pools and ponds in which, in addition to methane, small amounts of hydrogen sulfide are formed.

4.2 Sequences of appearance of forms, colors and smells

4.2.1 Degradation processes in the presence of air (Type 1a, b)

When considering our own breathing rhythm we can become aware of the fact that our bodily life continuously requires the transforming activity of the respiratory exchange with the open atmosphere. As in respiration, every life process proceeds in a rhythm that links outside and inside: during inhalation outer potential is transformed into the activity of the organism itself. Then the necessity arises to exhale – that is to get rid of something useless – followed again by the desire to inhale refreshing air from outside. In the natural life process of fading and degradation we are dealing with combinations of process conditions that correspond to these experiences. The 'spiritualizing' activities that disintegrate the complex plant residues are linked to the atmosphere in such a way that they give rise to specific environmental conditions. We often notice this in localities where access of air has been excluded and bad smells have accumulated. When we establish a well aerated degradation experiment, we live in the mood of a breathing and freeing connection of the transforming activities of the world. And we ensure that there is a neutral reaction (no acidification) and a continuously well mixed medium.

Conversion of forms: Initially the leaves and parts of them are present in fresh native form; the medium is slightly grayish, turbid from the inoculum. Gradually the leaves become soft, limp and lose their form. Then they disintegrate into smaller and smaller pieces causing an increasing turbidity and only the finest fibers remain. The leaf substance disappears in the liquid medium through its transforming activity. Correspondingly in our minds the species' specific pictures of form are gradually modified and trans-

nology.

4.1 Environmental conditions and a brief description of five types of degradation process

The most important distinction of different types of degradation process is given by the presence or absence of air (type 1 or types 2 to 5). Only if atmospheric oxygen is involved will all naturally formed organic compounds be completely degradable. In particular, the carbon bound in structure-forming substances will be released as gaseous carbon dioxide. Also, fully oxidized nitrogen and sulfur compounds are formed and all minerals are liberated in the oxidized state (type 1a, 1b). This way new plant root environment is potentially formed by decomposition processes dependent on oxygen. But whilst in the dark all transformations gradually cease (type 1b), in the light new plant growth processes set in that are perceptible through an increasing green coloration. Of course, in the liquid medium of the bottle cultures plant life remains restricted to the growth of cyanobacteria and micro-algae. Under corresponding conditions in nature, water-weeds and waterside plants develop in addition to algae.

Waste water and sewage treatment facilities often use open pond systems for the terminal purification of purified waste water. In such ponds waterside plants develop luxuriously and assimilate the minerals that are liberated in waste degradation. In specially arranged small scale sewage treatment systems with waterside plants (e.g. Phragmites, Juncus, Thypha) microbial degradation processes occur in the close vicinity of the plant roots.

Type 2 to 5: In the absence of atmospheric oxygen all degradation processes proceed more or less incompletely and finally suffocate themselves. Here we deal with conditions under which only bacterial transformations can take place. All higher forms of life cannot survive these conditions. Not until atmospheric oxygen gets access to the suffocated processes can transformation resume and gradually establish the conditions for new plant development.

Type 2: With fresh plant material, an acid fermentation starts in the bottle culture without additional inoculation or buffering of the medium. Lactic and acetic acids are formed by fermentation and they lower the pH so much (pH 3.5) that further degradation is inhibited and the plant material becomes acid-preserved. Transformations of this kind are used for the production of silage (from grass, white beet leaves, corn), sauerkraut and pickled gherkin. In nature similar conditions arise only in upland moors where the peat moss is preserved as peat by brown humic acids in the waterlogged oxygen free peat below the green surface layers.

Type 3: In the bottle cultures with freshwater of low sulfate content, an extensive but incomplete decomposition of plant residues occurs at neutral to weakly alkaline pH values. Instead of oxygen being reduced to water, carbon dioxide is reduced to gaseous methane (marsh gas, bio-gas, sewer gas) which escapes to the atmosphere. Marsh gas has a very high calorific value as it contains 60 - 80% methane. In order to obtain biogas from agricultural plant residues, large farms sometimes make use of this process in tall anoxic digestion tanks. At municipal sewage treatment plants sewer gas is obtained by fermentation of raw sewage sludge in huge sludge digesters. Further treatment and dis-

Oxidative stress has been shown to be a hallmark of prion diseases (Guentchev et al., 2000; Wong et al., 2001b; Sassoon, 2002). Numerous cell culture experiments have shown that infection with PrP^{Sc} causes oxidative stress and that addition of antioxidant molecules can alleviate the toxic effects of PrP^{Sc} to some extent (Brown et al., 1996; Perovic et al., 1997; Milhavet et al., 2000). Thus it is clear that oxidative damage is involved in the neurodegenerative mechanisms in prion diseases.

Co-incident with the appearance of PrP^{sc} in the brains of prion disease victims is a process called 'gliosis', which involves the activation of large numbers of cells called microglia. Various recent studies have shown the involvement of these cells in the prion disease process (Brown, 2001; Brown and Sassoon, 2003). The microglia mediate an inflammatory response in the nervous system and produce free radicals as part of their protective mechanism. Microglia usually exist as resting or quiescent cells that are exquisitely sensitive to pathological changes. Resting microglia are readily transformed into an activated state. They sense and respond to neuronal damage or unwanted particles, which they remove by phagocytosis acting very much like the white blood cells in the mammalian immune system. PrP^{sc} accumulation activates microglia because they recognize it as foreign particles. In the process of activation, the microglia produce free radicals. However, the surrounding neuronal cells are no longer able to clear the free radicals effectively because their antioxidant function has been compromised by the inactivation of PrP^{c} .

This model of prion-mediated degeneration involves two concurrent effects: an initial reduction in neuronal resistance to oxidative stress by the inactivation of the antioxidant PrP^C, (through conversion to PrP^{sc}) and a concommitant increase in reactive oxygen species produced by microglia that are responding to the PrP^{sc} accumulation.

Uric acid is an antioxidant A living organism will always try to respond to a detrimental process by trying to restore balance in the system. Thus in prion infected brains we might reasonably expect there to be a response to the loss of antioxidant function by PrP^{C} inactivation. One might predict that the brain will attempt to increase the production of other antioxidants as a countermeasure. Antioxidants include enzymes such as superoxide dismutase and PrP^{C} , certain vitamins, metal ions and uric acid. Uric acid is generated in mammals as a product of purine metabolism. Now we can return to the original question: does uric acid accumulate in the brains of BSE positive cattle? As an antioxidant, uric acid might be secreted in greater quantities in response to the loss of PrP^{C} . The brain may be using it to 'take over' the function of PrP^{C} .

Methods

Uric acid was detected in body tissues by two different methods: high performance liquid chromatography (HPLC) combined with electrochemical detection (ECD) (Yoshiura and Iriyama, 1986) and enzymatic assay (Amplex Red uric acid detection kit) (Kayamori et al., 1994; Mohanty et al., 1997; Nakaminami et al., 1999). HPLC with ECD is the more sensitive and accurate method and is most suitable for detecting uric acid in body fluids. Infected and age matched uninfected brain tissue (frontal lobe) and cerebrospinal fluid (CSF) were obtained from Central Veterinary Laboratory, Weybridge, U.K.

Cell culture experiments were designed to observe the behaviour of brain tissue cells *in vitro*. Tissues were initially trypsinised in 0.05% trypsin (Sigma) and the cells cultured on Dulbecco's minimal essential medium (Gibco) in 75 cm² Falcon culture flasks. The medium was usually supplemented with 10% foetal calf serum (Sigma) and 1% antibiotic/ antimycotic solution. Cultures were incubated at 37°C with 7% CO₂ for up to 14 days.

Results

The data presented below show that there is a significant, though not great, increase in uric acid in the brain tissue from BSE positive cows (Fig. 1). In contrast, the uric acid concentration in the CSF is lower in BSE infected cows (Fig. 2).



Figure 1. Unc acid in brain tissue (frontal lobe) from 19 cows with BSE (BSE+) and 19 age matched controls (BSE-). Error bars are presented as standard errors on the mean (SEM) on all graphs. e 2. Uric acid in cerebrospinal fluid (CSF) from 7 cows with BSE (BSE+) and 8 age matched controls (BSE-).

These results can be explained as follows. The increase in brain uric acid probably occurs in response to the loss of PrP^{C} antioxidant function. The antioxidant function of PrP^{C} involves a particular reaction with the superoxide radical in which PrP^{C} acts like a superoxide dismutase enzyme. A copper ion found in PrP^{C} and other superoxide dismutase enzymes is involved in the reaction, which inactivates the superoxide radical (Hough et al., 2000; Brown and Sassoon, 2002). The reaction can be represented as follows:

 $\begin{array}{l} \text{SOD-Cu}^{2+} + \text{O}_2 \xrightarrow{\cdot} \rightarrow \text{SOD-Cu}^+ + \text{O}_2 \\ \text{SOD-Cu}^+ + \text{O}_2 \xrightarrow{\cdot} + 2\text{H}^+ \rightarrow \text{SOD-Cu}^{2+} + \text{H}_2\text{O}_2 \end{array}$

It is clear that if PrP^C activity breaks down, superoxide radicals will accumulate unless inactivated in some other way. Excess superoxide ions are able to react with another radical called nitric oxide to form the peroxynitrite ion, a primary agent of neuronal damage.

 $NO + O_2^{-} \rightarrow ONOO^{-}$

of aquatic habitats. In soil, the solid mineral and plant components allow the formation of an extremely heterogeneous crumb structure which is interspersed by air and water. This is the habitat of a richly differentiated community of soil animals (e.g. insect larvae, wood-lice, Collembolae, Diplopoda, Polychaeta). The animals are active not only in the decomposition of plant residues but also in mixing and ripening the humus soil that becomes new plant root surroundings. Fungi and streptomycetes are the specific soil inhabitants among the micro-organisms (Pfennig 1984a, 1995); they grow on and through the solid substrates of the soil crumbs. To some extent we can compare the transformation processes of plant residues in soil with those in a compost heap, although the latter is a special organ with its own development taken care of by man. (Bockemühl 1978).

Compared with soil, the conditions for transformation processes in water (aquatic habitats) are very different. Water penetrated by light and in a state of more or less fluid movement incorporates all soluble substances homogeneously. Insoluble materials sink down to the sediment. In its upper layers containing oxygen, a rich community of small animals participates in the disintegration of plant residues. Among the microbes, bacteria are by far the most important terminal disintegrators of organic substances in water and sediment. Fungi by comparison are of minor importance. Because of the dissolving and mixing properties of water, transformation processes can be studied particularly well in aquatic habitats. Also, the various types of degradation process that we find under comparable environmental conditions in nature can be crudely imitated in the laboratory by the use of large liquid cultures.

4 Bacterial life processes in water

In an earlier publication (Pfennig 1988) a multiply replicated large scale experiment was described in which the degradation of plant material (green lettuce, pea pulp) in liquid cultures was compared under five typical sets of environmental conditions. Culture vessels were 10- and 20-litre bottles which were filled with 101 liquid. The 20-l bottles (with 101 liquid) were used for the aerated degradation experiments of the types 1a and 1b. The 10-1 free headspaces were provided for the contraction of foam that arose from aeration with aquarium pumps. This was unnecessary in the case of the anoxic vessels of the experiments of types 2 to 5. All vessels were stirred magnetically and checked daily over four weeks. Except for type 2, the pH of all vessels was adjusted daily to 7.0 to 7.5. The degradation processes were started with an active universal inoculation material (activated sludge and ditch mud) which had the potential for almost any kind of microbial transformation process. In nature this presents no problem, as microbes are widely distributed in the biosphere by water, wind and animals. The restriction to plant material appears justified, as its transformation is quantitatively most significant in nature. The transformation of animal body substances (alive or dead) is mostly started by carnivorous animals and results in other kinds of microbial processes and end products that will be mentioned later.

Firstly we describe the importance of the environmental conditions for the development of the various types of degradation process. This is done for the bottle culture experiment as well as for similar processes in aquatic habitats and in environmental techapparent by the fact that they are able to burn with a flame that gives light and warmth.

Corresponding to the magnification of the microscope through which microorganisms become visible we can characterize the metabolic process of a laboratory culture in nutrient medium as a 'physiological', or better still a 'processual magnification' (Pfennig 1988). This way we get to know the special metabolic capacities of individual microbial species which cannot be established in the context of natural degradation processes. With this approach, experimental microbiology can be compared with organic chemistry and biochemistry which also study the smallest partial processes. The appropriate way of working in these fields can be described as an indirect causation. This means that we only have to establish the conditions under which a given transformation will take place. In microbiology these conditions include an inoculation with soil, surface water or a microbial culture (process germs) in order to transfer the degradation activity.

As the nature and development of a transformation process are entirely dependent on the conditions established, we have to consider them carefully. They represent the specific configuration of environmental qualities that we experience as a mood or an atmosphere and they set the general tone of the sequence of appearances of the subsequent transformation process.

It is common practice in microbiology to represent the degradation process of a microbial culture by the sequence of structural formulae of the established intermediate and end products. In this way one sticks to objective representations and adds at best some conspicuous qualities.

However, in order to extend chemistry and microbiology it is essential to complement it by following the sequence of appearances of a process with inner participation and continued thought activity. The whole process will thus become one of inner perception. With this we follow indications given by Steiner (1923; see Bockemühl 1977, 1996, 2000).

3 Basic types of microbial life processes

To date, biochemical studies of the metabolic capacities of microbial pure cultures have revealed a vast number of different substrate transformations. There is hardly any organic substance of natural origin that cannot be degraded by microbes. But degradation processes by natural populations of microbes are known to proceed differently from what one would expect theoretically on the basis of the results of pure culture studies (Pfennig 1984a). The mass of known partial processes represents the other extreme to the largely interlinked natural transformation processes. Between these extremes there exist characteristic types of process involving microbial degradation of plant residues under certain kinds of environmental conditions. We will consider these as particular types of processes as we find them with modifications repeatedly in both nature and man-made process situations.

3.1 Soil and water

At first we have basically to differentiate between transformation processes proceeding on the moist surface soil below the vegetation and those occurring in water and sediments Both superoxide and nitric oxide levels were found to be elevated in the brains of scrapie-infected animals (Wong et al., 2001b) Uric acid is involved in neuroprotection because it acts as a direct scavenger of peroxynitrite (Hooper et al., 2000) or of secondary radicals resulting from the reaction of peroxynitrite with CO_2 (Squadrito et al., 2000).

Thus uric acid in BSE cow brains is increased because the tissue is responding to increased oxidative stress. Uric acid reacts with some of the excess free radicals and in the reaction is reduced to allantoin. The remaining uric acid and allantoin then are washed from the brain tissue into the CSF. This is why there is apparently less uric acid and more allantoin (data not shown) in the CSFs of BSE positive cows.

I was interested to discover which cells in the brain were responsible for the production of uric acid. Uric acid secretion by microglia, astrocytes and control (F21) cells was measured in cell cultures (Fig. 3). The microglia were the cells that secreted most uric acid. As already mentioned, microglia were also the cells producing the free radicals. Thus on sensing an increase in oxidative stress, the microglia themselves attempt to compensate for the potential damage to the brain tissue.



Figure 3. Secreted uric acid (UA) measured in cell cultures using the Amplex Red uric acid assay. F21 represents the control cell line. Astro = astrocytes. uglia = microglia. Microglia produce and secrete significantly more uric acid than either astrocytes or control cells. They are also most probably the primary source of uric acid production in living brain tissue.

The brain therefore reacts to the loss of PrP^C and increase in PrP^{Sc} in quite dramatic ways. It increases the number of microglia in order to try to remove PrP^{Sc} and dead cells but because microglia also produce free radicals as part of their protective mechanism there is an overall increase in oxidative stress. Consequently, microglia attempt to counter the situation by secreting more uric acid. My experiments have shown that uric acid increases in the brains of BSE positive cows even before symptoms appear (data not shown). It usually takes several years for an infected cow to demonstrate disease symptoms. The increase in uric acid, however, is detected earlier than symptoms. Thus the brain senses

and responds to changes induced by PrP^{sc} infection quite early on. Because symptoms eventually do appear, it shows that the protective mechanisms that come into operation are insufficient to cure an animal from the infection. Eventually they too become redundant.

I wondered if the protective mechanisms of uric acid could be demonstrated in a cell culture experiment. Fig 4. shows that indeed uric acid concentrations up to 100 mM can protect cells against abnormal PrP (in the experiments a synthetic peptide called PrP106-126, equivalent to PrPSc was used).



Figure 4. Uric acid attenuates prion peptide (PrP106-126) induced cell death. Uric acid concentrations between $20 \,\mu\text{M}$ and $100 \,\mu\text{M}$ significantly reduced neuronal cell death as compared to cells treated with peptide alone. At higher concentrations (around 1 mM and above) uric acid can enhance cell death.

Uric acid can cause cell death at high concentrations of 1 mM and above. Since Rudolf Steiner suggested that uric acid itself might be the 'cause' of the madness in cows, I considered the possibility that increased uric acid levels in infected brains might be the ultimate cause of neurodegeneration. On the basis of cell culture experiments, I had to conclude that this was unlikely. Firstly, the uric acid levels in brain tissue never accumulate to the neurotoxic levels I used in culture and secondly, I believe that it is far more likely that most of the neuronal death is caused by free radical damage.

Discussion

In conclusion I can give the following answers to the three questions I posed at the outset:

(1) Is the disease described by Steiner the same as BSE? Clearly it is not exactly the same. The whole process of prion disease is much more complex. I have tried to demonstrate that there is no single cause but that a number of processes have to

plants has set in, the colors become unattractive: the greens turn dull with yellowishbrown and grayish tints. Except for a few stalks, all forms become limp. When the vegetation begins to fade away we hardly notice the individual plants anymore. The initial richness of colors and forms that appealed to our sense of beauty finally changes into brown crumbs of soil. What has become withered and dead is now decomposed and feeds into an invisible but very active life process which proceeds in a direction counter to growth. It comes to rest as humus; native soil that represents a fertile environment for new root growth.

The process of soil formation from plant residues comprises the capacity for an unimaginable number of different transformation and degradation processes that depend on the continuity of the invisible living microbes, the process germs. Therefore, humus-rich soil represents a seemingly resting, but in fact active, labile state of chaos; not as a visible form like the plant but rather comparable to the resting state of seed or bud in higher plants. However, the direction of activity is reversed: under proper conditions the seed is ready to grow out of its state of chaos, with a wide range of possibilities for specific forms that differentiate themselves outwardly from within. But the corresponding stage of the soil has reached a peak of universal formation; it is continually ready to renounce itself and serve plant growth. At this stage the soil is open to its environment and has substances at its disposal so that each plant can find what it needs for its growth.

It is characteristic of all life processes in this field that they proceed imperceptibly. Only when disturbances occur do we often become aware that life processes have been 'quietly' serving; such disturbances are therefore connected with awakening and consciousness.

2 Processes in experimental microbiology

2.1 From natural process to laboratory culture

We can look at the development of microbiology during the past 130 years as an awakening to the transformation processes in soil and water. In order to get to know these processes together with their participating microorganisms, many different analytical methods in experimental microbiology were developed (e.g. Winogradsky 1949). Above all, pure cultures of bacteria and fungi were isolated and studied in the laboratory with respect to their metabolic capacities. In this way we came to know even the smallest component processes that might contribute to the overall process of transformation in nature (Pfennig 1984a, 1988, Schlegel 1992). Instead of using complex material formed originally by the plants themselves, individual pure organic substances (e.g. carbohydrates, fatty or organic acids, aliphatic or aromatic hydrocarbons, amino acids, purine or pyrimidine bases) together with minerals, were applied to culture media. These were inoculated under specific conditions (e.g. oxic or anoxic; acidic, neutral or alkaline). In this way it was possible to enrich and isolate particular degradation processes and their associated microorganisms in pure cultures. The organic substances contribute the 'chemical potency', the microorganisms the degradation activity that actualizes the potency. The chemical potency originates in the power of sunlight which gives rise to the synthesis of all plant substances. This unique cosmic-earthly origin of all organic substances becomes

Microbial processes and plant life - a key to a chemistry of life

Norbert Pfennig and Jochen Bockemühl

Summary

Like the growth and decay of plants, microbial decomposition processes belong to the life of the earth. Microbial transformations are characterized to show their position in the context of the earth in general and in relation to plant life.

Microbial decomposition processes of plant residues are differentiated according to the very different environmental conditions in which they occur. Two basic types are given in soil and water. Large scale experiments are described in which five different types of decomposition processes in water were studied: one under oxic conditions and four under various anoxic conditions. In each case the sequence of changes in appearance of shape, color and smell was described. The transformations were followed with inner participation. So the way the reaction conditions are arranged is experienced as the mood or atmosphere in which the entire process takes place.

Chemistry and biochemistry are concerned with the transformations of substances. These are consequently in a temporal and environmental context with other substances. For reactions to take place, chemical as well as microbial processes are initiated by establishing the proper reaction conditions. It appears appropriate to recognize microbial life processes in soil and water as part of a chemistry of life.

In contrast to laboratory experiments, microbial processes in nature are not limited in space and time as they proceed in the context of life in the biosphere of the earth. Plant development begins from the seed: roots grow down into the soil and the shoot above the ground grows and differentiates into special forms, colors and scents. The plant pheno-type becomes the expression of the specific nature of the plant. In contrast, the specific nature of micro-organisms, as process germs, gives rise to new environments. This is finally considered from the imaginative point of view which takes into account the relationship between inside and outside with respect to the earth: plant development above ground and microbial life processes in the soil are recognized as interconnected and complementary life processes.

1 Introduction

When the warmth and radiance of the sun increase each year we look forward to the emergence and unfolding of flowers. At that time the various sappy green colors of the leaves covering the ground are still conspicuous. Particularly striking are the shining blooms in white, pink, yellow, orange, blue and violet. The red hues appear somewhat later when we have become accustomed to the other colors.

What appeals to us is the splendor and beauty coming to prominence in all vegetation. Who at this time would think of the inconspicuous roots that are active in the hidden depths of the soil?

Later in the year it is quite different. When the withering and seed formation of the

operate for disease to come about. Uric acid is most probably not the reason for the 'madness' in BSE, but then neither is the presence of PrP^{s_c} the sole cause. The disease situation seems to arise from a number of co-ordinated processes

- (2) Does uric acid accumulate in the brains of cattle with BSE? Uric acid does accumulate in the brain tissue of BSE positive cows and, ironically, appears to play a protective function.
- (3) Can uric acid cause neurodegeneration as seen in BSE? Though I have stated that it probably does not play a role in neurodegeneration at the detected concentrations, it is impossible to say what the long-term effects of increased brain uric acid might be.

Out of necessity, I have presented a somewhat simplistic explanation of the processes taking place inside the brain in response to PrP^{C} inactivation and $PrP^{s_{c}}$ accumulation. It should not be assumed that uric acid is by any means the only antioxidant that functions in the nervous system. Neither should it be assumed that PrP^{C} is the only superoxide dismutase enzyme in cells. This is certainly not the case. There are several other well-characterized enzymes with this particular function. However, prion infection does ultimately disrupt the system and overrides all protective mechanisms sufficiently to create the perceivable disease situation several years after initial exposure.

It is, perhaps, also important to keep in mind an idea from anthroposophical literature that 'whenever lower organisms find suitable soil in the human (or animal *J.S.*) frame for development, that soil has been made suitable by the real primary causes of the disease.' (Steiner, 1920: GA 312). This quotation, though referring at the time to bacteria and human beings, might also be applied to viruses and prions. The suggestion is that an organism succumbing to an infection must already be somehow predisposed to it. This can be demonstrated. In our studies, not every experimental cow infected with BSE actually succumbs to the disease. Some appear more resistant than others. It is possible that individuals succumbing more quickly are not able to initiate a sufficient antioxidant response at the outset and so do not stave off the neurodegenerative processes as long as do animals with a stronger protective response. I am now collecting experimental data that supports this hypothesis.

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