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Solid State Fermentation and Food Processing: A Short Review

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Abstract

History of Solid State Fermentation (SSF) is very well known to everyone in the field of pharmaceuticals and food processing. It is widely applied to producing several enzymes, organic acids, flavoring compounds etc., which must be extracted and purified and then used in different products. However, there are very few scattered reports, reviewing SSF as an integral part of food processing. Today it plays a very important role in producing good quality, consumer acceptable and safe processed food items, which is essential for doing business on a global scale. There are several processed food produced traditionally using SSF in the unorganized sectors in every region, but then these are mostly in small scale and to cater the need of that particular community. In this article two of these products (produced in the organized sectors) have been dealt with in detail, along with microbial spoilage and disease causing organism using food as the vehicle of transmission.

Keywords: Bread; Cheese; Intoxicants; Spoilage; Microorganisms; Preservation

Introduction

Solid State Fermentation (SSF) is a fermentation method used by several industries like the pharmaceuticals, food, textile etc., to produce metabolites of microorganisms using a solid support in place of liquid medium. The support used is especially grain brans, deoiled oil seed cakes and other substances alike. The main advantage of such methods is that it produces a minimum amount of waste and liquid effluent thus not very damaging to the environment [1]. Solid state fermentation (SSF) has been used in food industry for various purposes like enzyme production, organic acid production, flavors, colors etc., [2,3]. However, there are many applications of such fermentation method directly in food processing. If one examines the role of SSF in food processing, then one would come across several instances, especially in traditional food processing the world over, like soy sauce preparation in the orient to bread making to cheese ripening in the west. Today, it has emerged as a profit center in food processing in the industries of organized sectors [4]. Initially, mostly fungi were used in such fermentation (as these microorganisms were considered to be very optimally active in very low water activity). Later, many bacterial species and yeasts were used to carry out such fermentation also [5]. It is essential to note that in food processing the cost of raw material is very important and if regulated properly, then it is possible to get acceptable, fermented foods at a very affordable cost. In many food processing industry, it is imperative to carry out highly regulated ageing, curing and ripening to get the right blend of flavors and essences in the processed foods. This is so because highly purified flavors or essences are added externally and then equilibrated in the product. However, in many cases, like in cheese ripening, it is possible to produce these during the processing itself like in fermentation process and often it does not require to be purified to a great extent [6]. Fermentation, often used to describe another very common food processing method, is pickling of fish, meat, eggs and and vegetable, to conserve the nutritional values in these items. It should be noted here that it is the lactic acid produced along with some amount of bacteriocins that help in preserving these foods by inhibiting most of the spoilage microorganisms like bacteria. At the same time it is rationale to justify such fermentative methods to produce lactic acid or bacteriocins is not by SSF, it is actually a submerged liquid fermentation (SLF). The fermentable carbohydrates are released from the substances, either by brining or by pressing like in Sauerkraut [7]. The lactic acid bacteria then convert these to lactic acid. However, pectinolytic and cellulolytic activities like sweating of coffee beans or cocoa beans or even curing of tea leaves can be termed as SSF.

Design parameters of SSF as applied to food processing

The major consideration in designing such a process is the kinetics of the fermentations involved. The constraint here is the amount of substrate utilized and the exact products formed. None of these are checked with very precisely, which is unlike the application of SSF in pharmaceuticals. However, with all these constraints, designing of SSF in food processing has and is taking place and is commercially viable too. Some of the important considerations are: first of all, the most important parameter-the growth pattern of the microorganisms i.e. biomass formation. There are certain mode of microbial growth in any fermentation especially in SSF, viz., the linear mode, the exponential mode, the logistic mode and the two phases of growth given by the following differential equations [8]:

Linear Mode: $dx/dt=K$

Logistic mode: $dx/dt=\mu x\ [1- (x/x_m)]$

Where K is the linear growth rate

Exponential mode: $dx/dt=\mu x$

Two phase Mode: $dx/dt=[\mu Le^{-k(t-ta)}]x,\ t>t_a$

Where μ is the specific growth rate

Where x is the biomass at time t; x_m is the maximum biomass; ta is the time when deceleration phase starts; k is the first order rate

constant and L is the ratio of specific growth rate of the deceleration phase and specific growth rate during the exponential phase. The graphs of these types of growth would be as shown in Figure 1.

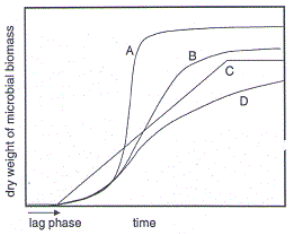

Figure 1: The various empirical kinetic profiles that have been described in solid-state fermentation systems: (A) exponential; (B) logistic; (C) linear; (D) fast- acceleration/slow-deceleration. In applying non-linear regression to experimental data zero time will typically be taken as the end of the lag- phase [8].

During most of SSF the microorganisms remain very closely associated with the solid substrates which hinder the measurement of the biomass. If the food which is undergoing fermentative processing is considered to be a bioreactor; then the two most important parameters that affect the process of SSF, are temperature and the water activity. Saucedo-Castaneda [9] derived the following equations for the logistic growth mode as:

$$\mu=[2.694 \times 1011 e(-70225/8.314T)]/[1+1.300 \times 1047 e(-283356/8.314T)]$$

$$x_m=-127.08+7.95(T-273)-0.016(T-273)^2-4.03\times10^{-3}(T-273)^3+4.73\times10^{-5}(T-273)^4$$

Where T is the temperature (K), how far these calculations will be valid in SSF mode of food processing, still remains to be answered. Thus it can be summarized that the rate of biomass formation is a function of T, a_w and Q (that is the physiological state of the cells) [10] and is expressed as:

$$dx/dt=f(T, a_w \text{ and } Q)$$

Another very important parameter which more than often is vital in determining the efficiency of any SSF is the death pattern of the concerned microorganisms. Again it must be remembered that the environmental factors are primarily responsible for the death of organisms. On the other hand, it is after the death of these microbes, the actual ageing or curing process starts where the important components of the processed foods (mostly flavors and essences) equilibrate with other components to give the required matured sensory and safety qualities. Just as it was difficult to quantitate the growing biomass in a food, similarly it is equally difficult to determine the death pattern. Death is defined as a permanent loss of the ability to grow. This definition does not imply that there will be a reduction in biomass. In case of fungi, sporulation may also be included in this definition of death. However, sometime dead cells undergo autolysis and liberate certain intracellular molecules which may be harmful or useful to the food undergoing SSF. Therefore, both these types of biomass are calculated in other SSF bioreactors using the equations specified by Sangsurasak, Nopharatana and Mitchell [11] and is as given below:

$$dx_v/dt=\mu_g x_v [1-(x_v+x_d)/x_m]-k_d x_v \text{ and}$$

$$dx_d/dt=k_d x_v$$

where x_v is the viable biomass x_d is the biomass of dead cells, μ_g is the actual specific growth rate and k_d is the first order death rate constant. In order to determine these two parameters (μ_g and k_d), Szewczyk and Myszka [12] suggested the following equation involving two Arrhenius type terms, in which μ_g and k_d are separately expressed as functions of temperature, as follows:

$$\mu_{obs}=\mu_g-k_d=\mu_{g0} \exp(-E_{ag}/RT)-k_{d0} \exp(-E_{ad}/RT)$$

Where μ_{obs} is the observed specific growth rate, μ_{g0} and k_{d0} are frequencies and E_{ag} and E_{ad} are activation energy factors for living and dead biomass respectively.

If one examines the yield of dry microbial biomass as a function of dry substrate (like carbohydrates) utilized, then it will be observed that dry microbial biomass produced will always be less than that of dry substrate utilized. It is essential to remember that both growth and no-growth related factors should be accounted for. The mathematical expressions for both substrate utilization and desired product formation rates are as:

$$dS/dt=[(-1/Y_{xS})\, dx/dt] - m_S x \text{ and}$$

$$dP/dt=Y_{Px}(dx/dt)+m_P x$$

where Y_{xS} and Y_{Px} are growth associated coefficients and m_S and m_P are non-growth (maintenance) associated coefficients. In some cases m_P is a non-growth associated production rate constant. These two equations will include different types of components. It is possible that P will sometime, be the energy released due to metabolism (often heat). Though it has been accepted that heat generated and released is directly proportional to the growth of microorganisms, actually it is not so as the maintenance heat when added to the growth related heat, it will be in excess [13]. The stationary phase and deceleration phase will show a decreased need for the substrate and then this decreased value need to be subtracted from the data when the measurement is a continuous procedure. The uptake of different substrate is not only dependent on the phase of growth but also on the affinity of the microorganism for that component, e.g., oxygen uptake will be different from that of saying glucose uptake, that is the specific uptake rate (q) for each component will be different for the same organism at each growth phase under the same environmental conditions of growth. Similarly the specific product formation rate will be dependent on the product, e.g., carbon dioxide will have a different specific formation rate at each growth phase which will be different from that of metabolic heat or from say a primary metabolite like ethanol or acetic acid etc. Likewise, decrease in maintenance activity (D) and m values will differ accordingly too.

These kinetic work-outs are mostly for bacteria, fungi and yeasts and not for protozoan. Of course it is needless to say that since viruses and similar organisms do not normally grow independently during food processing, these will not be discussed. However, it is important to note that certain viruses present in dormant conditions (like the lysogenic phages in fermenting bacteria or some similar viruses found in fungi and in yeasts) can affect the SSF during food processing. Such interferences are not possible to be modeled as these viruses cannot be easily quantitated in any culture, due to their random distribution in a given microbial population.

Types of food processing by SSF

There are two types of food processing carried out by SSF.

1. Processed foods as desired.
2. Unsafe or spoiled or undesired processed foods.

In the first type the two important processed foods that will be discussed will be the bread making by fermenting dough by yeasts and the second one is maturation of certain varieties of cheese. Note the Indian fermented food-idli is not an SSF, it Submerged Liquid Fermentation (SLF) with a high pulp density. Similarly, the soy sauce fermentation is actually a high salt containing submerged liquid fermentation, though the pulp density is fairly moderate.

SSF of dough for bread making

There are several reports as how the environmental factors like the temperature and aw affect the fermentation pattern of the dough. It should be remembered that fermentation of dough by *Saccharomyces cerevisiae* is very similar to that of sugar fermentation by the organism. The only difference is that this is an aerobic SSF where the end product is carbon dioxide. The next substrate is oxygen which is from the trapped air in the dough during the process of kneading. During the first three hours of fermentation the osmotic pressure increases by 23% and the yeast prefers to utilize glucose over fructose [14]. Further increase in time would lead to rapid exhaustion of oxygen and the yeast will resort to alcoholic fermentation which is deleterious for the product (bread) as it will severely affect the sponginess and flavor. Practically, it is not possible to measure the biomass developing nor the amount of carbon dioxide generated as a function of time. This is measured by noting the leavening of the dough. The lag phase will describe the carbon dioxide trying to reach the air nuclei and then diffuse from the dough. During this phase there will not be significant increase in size of the dough. Later it will be seen by the increase in size of the dough as the fermentation proceeds. The next important factor to be considered and found to be closely associated with the fermentation process is dough rheology, which is the texture of the final bread and that of the crumb. It was seen that as the yeast is growing in the dough matrix, it is producing carbon dioxide which is responsible for a type of cauchy strain on the chief protein of the wheat flour i.e. gluten. It must be remembered that gluten is a highly solvable protein. Normally the leavening process is restricted to 120 minutes only as further solvation of gluten would produce bread with very poor spongy texture. This is very well seen from Figure 2 [15], where Young Modulus (E) of the dough decreases with increase in fermentation time. Initially it has a linear relationship with the fermentation. This is best explained by the Maxwell model [16] where the relaxation force (F) at high values of time (t) and the equilibrium force (Fe) are related by the equation:

$$F(t)=F_e+A_1 \exp(-t/\lambda_1)+A_2 \exp(-t/\lambda_2)$$

A1 and A2 are initial values of force of the Maxwell elements and λ_1 and λ_2 are relaxation time for dough. It has been observed [17] that the elasticity of dough increases with Fe i.e with higher values of Fe the elasticity is high where as it was noted that at lower values of Fe there was increase viscosity of the dough instead of elasticity. It was also observed that with lower $\lambda 1$ there is initial stress decay, whereas λ_2 indicates stabilization of the gluten structure of the dough i.e. it forms a uniform viscoelastic mass for the air bubbles, starch molecules and other materials. As far as the rheological properties of bread crumb is concerned, it has been found to be anisotropic. Initially during the first 30 to 50 minutes of fermentation, the Young Modulus decrease by 3 to 5 folds resulting in good crumb properties. However, after 120 minutes the gluten gets highly solvated and the elastic properties are virtually totally replaced by viscous properties (with very small values of λ_1 and λ_2) and it results in very poor crumb qualities.

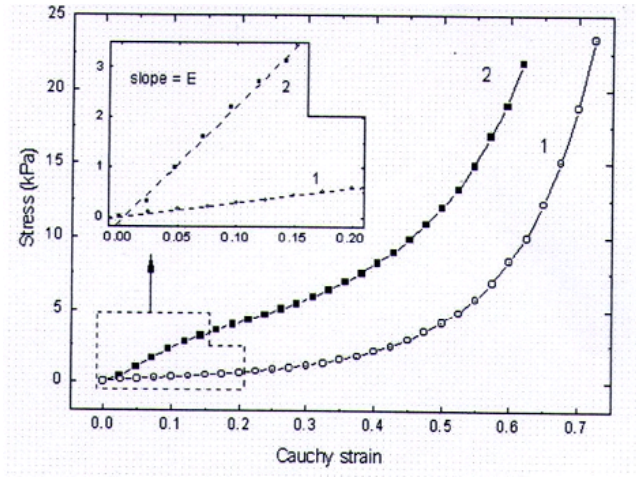

Figure 2: Compression curves for dough after 120 minutes of fermentation (curve 1) and bread crumb obtained from this dough (curve 2) [15].

This implies that the fermentation time should be 120 minutes maximum. These observations have been recorded while working with white bread. Today there is a wide variety of bread than the standard white bread. One of the important of such types is the multigrain bread where using the bleached flour of wheat as the base, there are different grain flours that are added in different proportions, like the sorghum flour, millet etc., These flours are not viscoelastic like the bleached and refined wheat flour. Fermentation of such dough no doubt shows leavening, but the end product has totally different characteristics, especially the crumb properties. Many a time these are flakier in crumb structure, yet retaining the soft and spongy nature to a large extent, thus making it acceptable to a restricted class of consumers. This dough does not follow the Maxwell Model as stated earlier. This is because though the values of λ_1 and λ_2 will be extremely small, they do not show the viscous properties so significantly. Not that these retain the elastic properties either but are more complex blend of several properties, each showing the separate flour properties of the specific grains added and in different quantities present along with the amount of shortening used to knead the dough.

SSF in cheese ripening

There are only 2 stages involved in cheese manufacturing process, where fermentation is involved. This is at the very first stage, where the pasteurized milk is fermented with *Lactococcus spp* to lower the pH for the next step of casein precipitation by rennet. This is a submerged liquid fermentation (SLF) which will not be discussed here. The last stage, which is called as the ripening or maturation of the cheese, involving either bacteria (mostly lactic acid bacteria, *Propionibacterium spp*) or soft fungi like *Penicillium* or some yeasts like *Debaromyces hansenii*, where the biomass should be maintained to the minimum and metabolite production should be optimum. The other stages like ageing and curing normally do not involve any microbial activity. Maturation is achieved by either reducing the water activity or brining or reducing oxygen tension. These are very time consuming process, running from a few months to a couple of years. It

involves some proteolytic activity, lipolytic activity and someime even saccharolytic activities too to produce gas like carbon dioxide and finally equilibrating the liberated substances to get the desired flavor, texture and appearance. There are very stray reports regarding the kinetics of these microbial processes. This is due to the fact that the process of maturation of cheese is very complex involving several but continuous steps each with its own kinetics. Though it has been observed that in cheese like feta cheese, the ripening process does not interfere with pH or mineral content but does show a significant increase in soluble nitrogen content [18]. Secondly, it must be remembered that in a matrix like that of cheese, whatever may be the preset environmental conditions seeking a particular maturation, the results obtained will always fall below the benchmark (may not be very significantly) because as the microbes are living and carrying on with the metabolic activities, they will create a micro niche around them which will be different from that of the surrounding. It is due to the fact that every organism wants to live in a stress free environment. This makes it difficult to study any kinetic of this sort of fermentation. That is the rationale that the best matured or ripened cheese is produced by manufacturers where the environmental factors has been preset or determined by trial and error methods and is being handed over from one generation to another. Earlier Kim et al [19] had tried to study the ripening process, wherein they divided the time factor in 2 stages: 1) that lasts for two to four weeks and here the bacterial cells undergo significant changes, but as the lactose gets depleted and the cells enter the death phase. During this phase enzymes would be produced which brings about considerable changes in the green cheese matrix. 2) This is a phase where exclusively different biochemical reactions are taking place and the products are trying to attain some sort of steady state condition to give the desired texture and flavor. If one examines the maturation of cheddar cheese as a study model, then the following first order reactions play vital roles. Some of these equations are as [20]:

During phase1: The bacterial growth is:

$dX/dt=[\mu_m LX/ (K_l+L)]-k_l X$

Where μ_m is the maximum specific growth rate, t is time, L is the amount of lactose, X is the biomass of cells, Kl is the rate constant and kl is the death rate constant.

Lactose utilization is given by:

$dL/dt=[-(\mu_m/Y_x)-\mu_{LA}]\ LX/(K_l+L)$

Where L is the lactose concentration, μLA is the specific rate of lactic acid formation and Y_x is the yield factor (biomass formed per unit lactose utilized).

Lactic acid formation is:

$dLA/dt=K_1\ [\ \mu_{LA} LX/(K_l+L)]$

where K_1 is the proportionality constant for lactose and lactic acid and LA is lactic acid.

Proteinase formation is:

$dP_1/dt=[.\alpha_1\mu_m LX/(K_l+L)]-k_1 P_1$

where P_1 is the proteinase concentration and α1 the reaction constant and k1 is proteinase destruction rate.

Extracellular dipeptidase formation is:

$dP_2/dt=\mu k_l X-k_2 P_2$

where k_2 is the dipeptidase destruction rate, P_2 is the concentration of dipeptidase and μ is the dipeptidase reaction constant.

Extracellular lipase formation is:

$dE_{Lp}/dt=\mu_{Lp}$ kl $X-k_3 E_{Lp}$

Where E_{Lp} is the concentration of extracellular lipase, k_3 is the E_{LP} destruction rate and μ_{Lp} is the extracellular lipase reaction constant.

It must be borne in mind that howsoever, it may appear simple to solve these first order equations, it is not so, because the fermentation is going on in a solid matrix like cheese and these enzymes or acids formed during maturation may not be all that evenly distributed. However, the transformed products of these enzymatic reactions can be estimated with an average value having the required standard deviation. Since the end products which are responsible for the desired flavor and texture are the consequences of these three enzymatic reactions, it becomes imperative to calculate the rate of transformation of the substrates or formation of products like free amino acids or fatty acids. This is the second phase of the maturation process. The necessary equations are as:

The rate of casein transformation is:

$dC/dt=-\ [\ V_f E_1 C/(K_C+ C)]$

Where V_f is the reaction velocity constant, K_c is reaction constant and C is the amount of casein transformed.

The rate at which amino acids liberated is:

$dA/dt=V_D\ E_2\ D/(K_D+D)$

Where V_D is the reaction velocity constant, K_D is reaction constant, D is the amount of dipeptides released and A is the amount of amino acids.

The rate at which dipeptides are released is

$dD/dt=-\ \zeta\ dC/dt - dC/\zeta dt$

Where ζ is scaling constant for proteolytic reactions and D is the amount of dipeptides.

The rate at which triglycerides are utilized is

$dT/dt=-V_T\ E_{LP}\ T/(K_T+T)$

Where VT is the reaction velocity constant, K_T is reaction constant and T is the amount of triglycerides utilized.

The rate at which fatty acids are released is

$dF/dt=-K_2\ (dT/dt)$

Where F is the amount of fatty acids released.

It is essential to note that in case of large volume of data processing, these differential equations must be suitably integrated. Secondly, these equations would give two categories of results, viz. a set of linear and another set of nonlinear results. If the tolerance value is set at >1 (which is often the case), then one would have to work with very restricted number of data to infer some sort of designing parameters. This is very well demonstrated by Sweatman et al. [20] in Figures 3-7. It can be very well seen from Figure 3, that what is supposed to be a growth curve of the organisms responsible for ripening is actually a death pattern. This is because when the precipitated casein is drained of its whey, most of the lactose comes as whey solids and very little remains in the coagulated casein for further processing. This is very well shown by Sweatman et al. [20] in Figure 4. Similarly as the

microorganisms are dying, the proteinase activity is limited as seen in figure 5 where the amino acids liberation should have gone on for 350 to 400 hours, ends in less than 50 hours. It is a similar result with the dipeptidase reaction too as seen in figure 6, where the release of dipeptides ends abruptly at 150 hrs. The exception is the lipolytic activity where in figure 7 it has been going on for 400 hours and it is predicted that the free fatty acids in the ripened cheese are the ones contributing to the flavor of cheddar cheese.

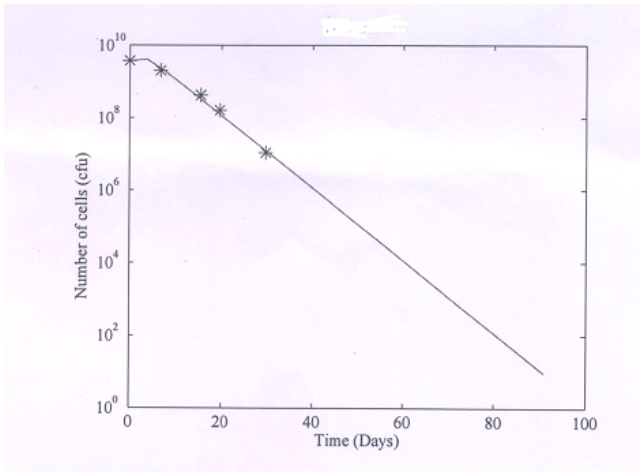

Figure 3: Bacterial growth pattern in very limiting substrate concentration of lactose. It is virtually a death pattern. The solid line shows the expected pattern and (*) shows the actual results [20].

Depectinolytic activity of cocoa and coffee beans

This is an essential SSF to get good quality coffee and cocoa beans and if carried out properly, then controlled roasting of the beans will bring out the best flavor and aroma. These are mostly made by the traditional method of burying the pectin containing pods in a pit and then covering it with some easily degradable agriculture waste like layers of banana leaves. Where in as the banana leaves undergo composting, it generates enough heat for the depectinolytic activity of the microorganisms.

The right type of maturity of the leaves to get the right incubation temperature in the heap, time of incubation and the maturity of the pods, are secrets (tried and tested thumb-rules) which are passed on from generation to generation in the farming community. There have been some attempts to study the transition of the microbial population in such a heap but no detailed kinetic study has been attempted. A study of some of the microbes, associated with such SSF carried out spontaneously, has been done by de Melo Periera et al. (2014) [21] revealed yeasts from the genera *Candida, Saccharomyces*, *Pichia* and *Hanseniaspora* were involved. Of these *Saccharomyces* spp were primarily responsible for maximum pectinolytic activities and *Pichia fermentans* gave the flavor acting compounds like ethyl acetate and isoamyl acetate. It was observed that the yeast population showed maximum growth by the 40th hour and then started declining. The study was carried out for 48 hrs only.

Similarly, Lefeber et al. [22] reported that *Lactobacillus plantarum* and *Lactobacillus fermentum* along with *Acetobacter pasteurianus* enhanced the flavor of cocoa beans, during SSF, using Pulp Simulation Media (PSM). However, there is no further report as how this has improved the cocoa bean sweating process under field conditions.

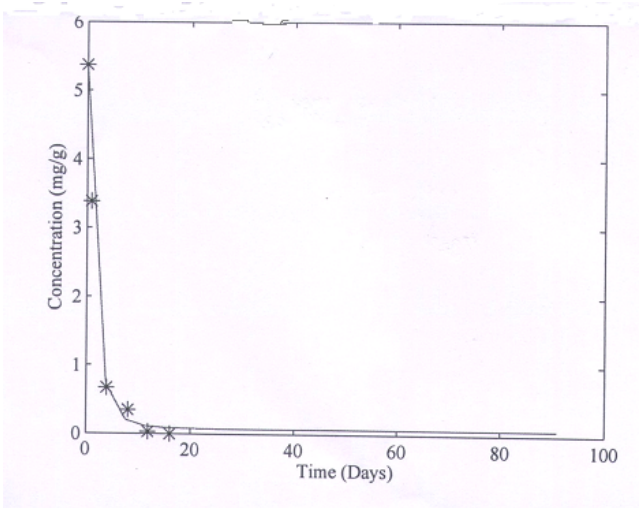

Figure 4: Lactose utilization curve. The solid line shows the expected pattern of utilization and (*) shows the actual results [20].

Curing of tea and tobacco leaves

There is tremendous amount of controversy over the curing process of tea and tobacco leaves. There some unpublished information that *Bacillus megaterium* and *Micrococcus condiscan* are involved in curing of both these types of leaves. However, there are no scientific published reports of such microbial action. Therefore, such curing process cannot be accepted today as some sort of SSF. It is believed that oxygen from the atmosphere and certain polyphenol oxidase present in the wet leaves do the curing job and imparts the requisite flavor and aroma.

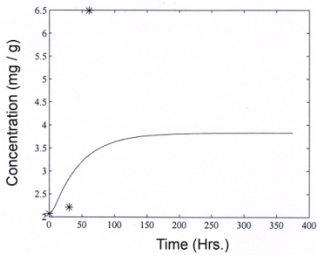

Figure 5: It is the pattern of free amino acid production after the action of proteinase on casein. The solid line shows the expected results and (*) shows the actual results [20].

SSF in spoilage of solid food items

Until now one has seen the positive effects of SSF in food processing and how different value added products have been made by applying this technique directly in food processing.

Spoilage is also a method of food processing but in a negative sense as it reduces the value of the food items and make it unfit for human consumption. The objective of studying the kinetics is to provide information as to how best it can be reduced if not prevented. The biggest problem is that nobody ever designed a spoilage process (as it happens naturally without any forewarning) and hence certain basic information will always be missing, which will be replaced with the probable theoretical assumptions. One such important information is the number of microorganisms that were present initially to bring about the spoilage (or what is called as the inoculum size). The major hurdle is that spoilage organisms have different degree of their capability to cause the biotransformation of the substrate, as these are natural contaminants. As pointed out earlier, if this inoculum size could be determined then it would make Hazard Analysis at Critical Control Points (HACCP) a much simpler job, since the points could be defined more precisely, through which these spoilage organisms could get access to the food material.

SSF of pathogens surviving and growing in different food

This has been well studied with dairy products like cheese which are fermented food basically with Lactic Acid Bacteria (LAB). Actually LAB inhibit the growth of other organisms by three means: 1) By producing lactic acid and other organic acids which lower the pH of the food; 2) By producing bacteriocins which are inhibitory to other microorganisms and 3) By producing H_2O_2 which are harmful to many microorganisms.

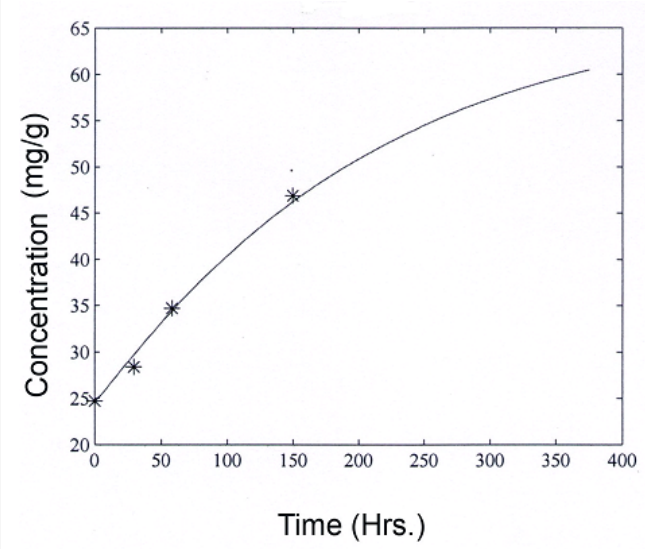

Figure 6: This shows the dipeptide production from the dipeptidase action. The solid line is the expected results and (*) it is the actual observation [20].

In spite of these sorts of natural preservatives, these foods are stored in low temperature (usually between 4°C to 5°C) where most of the human pathogens get inhibited and slowly cease to grow. In addition, cheese are often protected by addition of external preservative (as per the rules of FDA of that region) and sometime coated with edible wax so that unwanted microorganisms are kept out. Actually these different types of preservatives act as stress factors for the growing microorganisms. On the other hand, one would see that cheese is a very good nutrient medium for the growth of different organisms as these contain growth factors like many essential amino acids (from partial digestion of the casein) and vitamins. Therefore, even in presence of various stress factors, the presence of growth factors enable pathogens to grow on such medium like cheese. Some of the examples of different pathogens capable of growing in cheese are the hemorrhagic colitis causing *E. coli*-O 157: H7. This organism is highly acid adaptive [23]. Another organism commonly found in other types of fermented milk like yogurt and not so common in cheese, has been reported to be found in products like spread cheese, is *Listeria monocytogenes*. This bacteria is not only acid resistant (as it is also a lactic acid producer), but also is a psychrotroph, i.e., it survives for more than 15 days or more at refrigeration temperature like 4°C [24]. Since cheese is not a source of probiotics, *L. monocytogenes* can adhere to the intestinal mucosal cells to cause infection which otherwise would have been prevented by lactic acid bacteria in probiotics. The most dreaded is *Staphylococcus aureus* (now *Micrococcus aureus*) which can adapt to acid environment and to high salt concentrations [25] and is capable of producing a highly potent heat stable exotoxin.

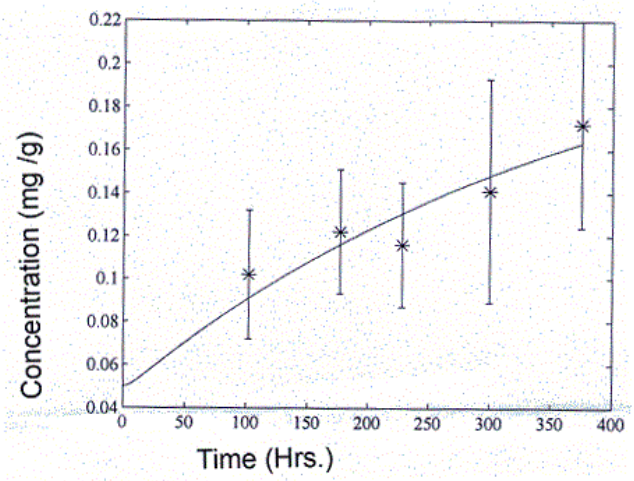

Figure 7: This is how the lipolytic action takes place. The solid line shows the expected fatty acid liberation and (*) shows the actual pattern of fatty acids released [20].

Another important disease caused by metabolites of microorganisms is carcinogenesis. Many times it has been observed that certain microorganisms capable of growing on solid food produces very potent carcinogens [26]. Fungi have been on top of the lists of such microorganism. Many fungi grow on solid food with minimum water activity and produce certain nitroso compounds which are carcinogens [27]. A substance known as roussin red methyl ester has been isolated from pickled vegetables where certain yeasts were growing and this compound is a known cancer promoting agent [28].

Conclusion

The importance of studying positive solid state fermentation (SSF) in food processing, is primarily aimed in optimization of parameters,

so that a suitable design can be achieved which will make a process more commercially viable. On the other hand, the negative SSFs are equally important. It would be very interesting to find reports of kinetics of such processes. This would go a long way in not only reducing spoilage of food (either processed or unprocessed) as it would precisely point out the critical points during processing, which can be a potential source of risks (hazards). These then can be monitored and checked appropriately. Since it is known that, quality management (quality control and quality assurance) is primarily associated with management of risks or hazards, the source of which are the several variables in the process. In SSFs, therefore, trying to define some of these would reduce their number and would definitely result not only in good quality, consumer acceptability and safe processed food products.

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