Single Cell Protein
Single cell protein

- The term single cell protein (SCP) refers to dead, dry cells of micro-organisms such as yeast, bacteria, fungi and algae which grow on different carbon sources. The name "single cell protein" was used for the first time by the M.I.T. professor Carol Wilson to give a better image than "microbial protein".
PURE CULTURE FERMENTATIONS
- industrial ethanol
- alcoholic beverages
- fermented foods
- pharmaceuticals
- acetone-butanol
- acetic acid
- single cell protein
- industrial enzymes
- biotech products (insulin, growth hormone)

http://www.agen.ufl.edu/~chyn/age2062/lect/lect_13/lect_13.htm
Table 1  Mass doubling time (S)

<table>
<thead>
<tr>
<th>Organism</th>
<th>Mass Doubling</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bacteria and yeast</td>
<td>10-120 min</td>
</tr>
<tr>
<td>Mold and algae</td>
<td>2-6 h</td>
</tr>
<tr>
<td>Grass and some plants</td>
<td>1-2 wk</td>
</tr>
<tr>
<td>Chickens</td>
<td>2-4 wk</td>
</tr>
<tr>
<td>Pigs</td>
<td>4-6 wk</td>
</tr>
<tr>
<td>Cattle</td>
<td>1-2 mo</td>
</tr>
<tr>
<td>People</td>
<td>0.2-0.5 yr</td>
</tr>
</tbody>
</table>

Table 2  Efficiency of protein production of several protein sources in 24 hours (16)

<table>
<thead>
<tr>
<th>Organism (1,000 kg)</th>
<th>Amount of protein</th>
</tr>
</thead>
<tbody>
<tr>
<td>Beef cattle</td>
<td>1.0 kg</td>
</tr>
<tr>
<td>Soybeans</td>
<td>10.0 kg</td>
</tr>
<tr>
<td>Yeast</td>
<td>100.0 tn</td>
</tr>
<tr>
<td>Bacteria</td>
<td>100x10,000,000 tn</td>
</tr>
</tbody>
</table>
Nutritional Value of SCP

For the assessment of the nutritional value of SCP, factors such as nutrient composition, amino acid profile, vitamin and nucleic acid content as well as palatability, allergies and gastrointestinal effects should be taken into consideration. Also long term feeding trials should be undertaken for toxicological effects and carcinogenesis.

Table 3
Average composition of the main groups of micro-organisms (% dry weight)

<table>
<thead>
<tr>
<th></th>
<th>Fungi</th>
<th>Algae</th>
<th>Yeasts</th>
<th>Bacteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein</td>
<td>30-45</td>
<td>40-60</td>
<td>45-55</td>
<td>50-65</td>
</tr>
<tr>
<td>Fat</td>
<td>2-8</td>
<td>7-20</td>
<td>2-6</td>
<td>1.5-3.0</td>
</tr>
<tr>
<td>Ash</td>
<td>9-14</td>
<td>8-10</td>
<td>5-9.5</td>
<td>3-7</td>
</tr>
<tr>
<td>Nucleic acids</td>
<td>7-10</td>
<td>3-8</td>
<td>6-12</td>
<td>8-12</td>
</tr>
</tbody>
</table>
Table 4
Essential amino acid content of the cell protein in comparison with several reference proteins (grams of amino acid per 100 grams of protein)

<table>
<thead>
<tr>
<th>Amino acid</th>
<th>Cellulomonas</th>
<th>Sacharomyces</th>
<th>Saccharomyces</th>
<th>Spiritulis</th>
<th>Penicillium</th>
<th>B.P. (SCP)</th>
<th>Wheat</th>
<th>Egg</th>
<th>Milk</th>
<th>Cow</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lysine</td>
<td>7.6</td>
<td>7.7</td>
<td>4.6</td>
<td>3.9</td>
<td>7.0</td>
<td>2.8</td>
<td>6.3</td>
<td>7.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Threonine</td>
<td>5.4</td>
<td>4.8</td>
<td>4.6</td>
<td>4.9</td>
<td>2.9</td>
<td>3.2</td>
<td>4.6</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Methionine</td>
<td>2.0</td>
<td>1.7</td>
<td>1.4</td>
<td>1.8</td>
<td>1.5</td>
<td>3.2</td>
<td>2.4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cysteine</td>
<td>2.0</td>
<td>1.0</td>
<td>0.4</td>
<td>2.5</td>
<td>2.4</td>
<td>3.2</td>
<td>2.4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tryptophane</td>
<td>5.3</td>
<td>4.6</td>
<td>6.0</td>
<td>4.5</td>
<td>3.3</td>
<td>6.8</td>
<td>6.4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Isoleucine</td>
<td>7.3</td>
<td>7.0</td>
<td>8.0</td>
<td>7.0</td>
<td>6.7</td>
<td>9.0</td>
<td>9.9</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leucine</td>
<td>7.1</td>
<td>5.3</td>
<td>6.5</td>
<td>5.4</td>
<td>4.4</td>
<td>7.4</td>
<td>6.9</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Valine</td>
<td>4.6</td>
<td>4.1</td>
<td>5.0</td>
<td>4.4</td>
<td>4.5</td>
<td>6.3</td>
<td>4.9</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>7.8</td>
<td>2.7</td>
<td></td>
<td>2.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Histidine</td>
<td>6.4</td>
<td>2.4</td>
<td></td>
<td>4.8</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

Microbiological proteins are deficient in the sulphur amino acids cysteine and methionine and require supplementation, while they exhibit better levels of lysine.
Other nutritional parameters which evaluate the quality of a given SCP are:
- the digestibility (D)
- the biological value (BV)
- the protein efficiency ratio (PER)
- the net protein utilisation (NPU)
The Problem of Nucleic Acids

About 70-80% of the total cell nitrogen is represented by amino acids while the rest occurs in nucleic acids. This concentration of nucleic acids is higher than other conventional proteins and is characteristic of all fast growing organisms.

The problem which occurs from the consumption of proteins with high concentration of nucleic acids (78-25 g/100 g protein dry weight) is the high level of uric acid in the blood, sometimes resulting in the disease gout. Uric acid is a product of purine metabolism. Most mammals, reptiles and molluscs possess the enzyme uricase, and the end product of purine metabolism is allantoin. Man, birds and some reptiles lack the enzyme uricase and the end product of purine degradation is uric acid.

The removal or reduction of nucleic acid content of various SCP's is achieved with one of the following treatments: chemical treatment with NaOH; treatment of cells with 10% NaCl; and thermal shock. These methods aim to reduce the RNA content from about 7% to 1% which is considered within acceptable levels.
The advantages of SCP over conventional protein sources are:
- the small doubling time of cells (td);
- the productivity of protein production form micro-organisms is greater than that of traditional proteins;
- it is independent of land and climate;
- it works on a continuous basis;
- it can be genetically controlled;
- it causes less pollution.

There are five factors that impair the usefulness of SCP:
- non digestible cell wall (mainly algae);
- high nucleic acid content;
- unacceptable coloration (mainly with algae);
- disagreeable flavour (part in algae and yeasts);
- cells should be killed before consumption.

Thus SCP is treated with various methods in order to:
kill the cells;
improve the digestibility;
reduce the nucleic acid content.
Flow chart for single-cell protein production

- Substrate
  - Fermentor
    - Nutrient
    - Filtration
    - Drying
      - Single-cell protein (SCP)
        - Submerged fermentation
        - Semisolid fermentation
http://www.agen.ufl.edu/~chyn/age2062/lect/lect_13/lect_13.htm
http://www.fao.org/docrep/X5738E/x5738e2w.gif
SCP from N. Alkanes

In the late 1950's, British Petroleum (BP) became interested in the growth of a microorganism in $C_{12} - C_{20}$ alkanes. This constitutes the wax fraction of gas oils for treating. Some crude oils contain up to 15% in wax, and these waxes must be removed since they make oil more viscous, precipitate out at low temperatures, block tubes etc.

BP uses two yeasts, *Candidor lipolytica* and *C. tropicalis* and built a 16,000 tons/year plant in Cap Lavera, France, and a 4,000 tons/year plant in England. The product produced was called "TOPRINA". In the UK the product "TOPRINA G" was a purer product while the one in France was not separated from alkanes. Both processes employed $NH_3$ as N-source and Mg ions to increase yields. No other carbon source was used. TOPRINA was tested for toxicity and carcinogenecity and was marketed as a replacement for fish meal in high protein feeds and as a replacement for skimmed milk powder in milk replacers.

Pigs fed on 30% TOPRINA in their diets showed less n-paraffins in their fat tissue than those fed on pasture. Based on this evidence the Italian government agreed to the use of TOPRINA in limited amounts and only for export. In 1977 Italy stopped the SCP production for alkanes altogether due to the increase in oil prices. The price of soya was more competitive. Now there is no factory which produces any petrochemical protein.
SCP from Methane

Methane is cheap, abundant and without the toxicity problems. Methane contains the most highly reduced form of carbon and consequently gives high cell yields relative to the amount of gas consumed. The general *Methylomonas* and *Methylococcus* have been recognised as utilising methane as a carbon source. The species which has been extensively studied is *Methylomonas methanica*. Nitrates or ammonium salts can serve as N-source.
The most important work in this field was carried out by Shell in England. The process involves methane oxidation by stable mixed cultures. These were
- a methane utilising G(-) rod;
- a Hyphomicrobium;
- two g(-) rods; *Acinetobacter* and *Flavobacterium*
This mixed culture was one of the best examples of symbiosis.

The process began in 1970 with a goal of producing 100,000 tn/year. In spring 1976, Shell stopped commercialisation and its development plans were indefinitely postponed. This decision was based on 3 factors:
the low price of soybeans & maize;
the potential of many countries for expanding existing protein sources;
the difficulty in applying Shell's sophisticated process in underdeveloped countries.
Production of single cell protein from cassava

The use of cassava as substrate for single cell protein has been investigated since the mid-1960s. Gray and Abou-El-Seoud (1966) grew some filamentous fungi on ground cassava roots, supplemented with ammonium chloride and corn steep liquor, to obtain biomass containing 13-24% crude protein.

*Candida utilis* fermented enzymatically hydrolyzed cassava in a submerged culture to produce a product containing 35% crude protein on a dry weight basis (Shrassen *et al.* 1970).

*Aspergillus fumigatus* 1-21 A fermented whole cassava in a nonaseptic continuous fermentation system to produce single cell protein containing 37% crude and 27% true proteins. This product was fed to rats and produced good growth responses.
*Rhodopseudomonas gelatinosa*, a photosynthetic bacterium, was cultivated on cassava starch medium under aerobic dark and anaerobic light conditions. The optimum temperature for growth was found to be 40°C with maximum growth rate and growth yield of 0.23h-1 and 0.40g cell/g starch and 0.13h-1 and 0.83g cell/g starch, respectively for the aerobic dark and anaerobic light conditions (Norparatharaporn *et al.* 1983).

Ghoul and Engasser (1983): enrichment of cassava by enzymatic hydrolysis and *Candida utilis* fermentation. In this process, cassava starch is first liquefied by thermostable $\alpha$-amylase and then saccharified by glucoamylase before fed-batch fermentation of the hydrolyzed cassava by *Candida utilis*. A product with 70g/L yeast concentration was obtained after 20 hours of fermentation at a maximal 50% biomass conversion, with 40% crude protein.

http://www.fao.org/Wairdocs/ILRI/x5458E/x5458e0d.htm
http://www.agen.ufl.edu/~chyn/age2062/lect/lect_13/lect_13.htm
The EversTech Process plant in Fresno, CA, owned by Cottonwood Creek Biosystems, takes in tankered grease and food processing wastes and generates single cell protein and dischargeable water.

The EversTech’s Process, is a biological system for the treatment of organic wastes. The process incorporates bio-augmentation, whereby specific bacterial formulations tailored to the waste are introduced into the system. The ET Process are optimized and controlled to generate a single cell protein as animal or aquaculture feed with a basic potential value of over $1 million per month to the ethanol plant, but many times that if supplied to an on-site fish farm, for example.
Crypto Power ™ Chlorella is a single cell, thin soft cell wall chlorella (Cryptomonadales, *Chlorella sorokiniana*) that is 95% digestible. Crypto Power ™ chlorella is one of the highest natural sources of chlorophyll and phycocyanin. Plus, it contains an abundant variety of micro nutrients not found in any other chlorella strain, including significant amounts of all natural alpha, beta and gamma PPARs. Peroxisome Proliferator Activated Receptors
SCP's Future Prospects

Today in most countries where market forces operate. SCP cannot compete with soya, alfalfa or fish meal. Mushroom production from lignocellulosics seems to be one economical and promising use for SCP. For future success of SCP, first, food technology problems have to be solved in order to make it similar to familiar foods and second, the production should compare favourably with other protein sources.

http://www.biopolitics.gr/HTML/PUBS/VOL1/isreali.htm