Introduction

The cultivation and processing of grain is probably one of the most important technological achievements on humanity's path to modern civilization. 10,000 years ago, the first grains were already being cultivated in the Middle East, leading to the development of the first advanced civilizations. Today, various types of grains have become staples worldwide and thus essential to the basic livelihood of every society. Among them, wheat is probably the most well-known and important grain. In addition to wheat, many other grains contribute significantly, both directly and indirectly, to securing our basic nutritional needs.

Rye

One such grain is rye, which, like almost all grains, comes from the family of sweet grasses (Poaceae = Gramineae). Due to its modest requirements in terms of soil quality (nutrient-poor and acidic soils), rye has taken on particular importance in Northern and Eastern Europe. In recent years in particular, this grain has attracted increasing research interest, in part because its high hemicellulose content (fiber) may be an important component in the prevention of colorectal cancer. In addition, its high concentration in amino acids is extremely interesting from a nutritional point of view. Aside from its purely medicinal and...
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HYDROVEG® R – Copying the Skin NMF
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nutritional importance, rye is of increasing importance as an alternative grain in sustainable agriculture (green fertilizer, soil improvement).

### Process

Both the cultivation of grains as well as the processes used to transform these plants into better food sources date from roughly the same period (circa 10,000 BC). Early on, mankind recognized how to modify many natural substances by means of microorganisms so as to make them available to us as foodstuffs.

The most well-known process is undoubtedly that of meat maturation, by which lactic acid bacteria and their proteases break down the muscle proteins so we can better digest and utilize them. Human beings were aware of how this process operates even when they were still hunters and gatherers. When they started to settle down, these processes were extended to grains and dairy products, making the production of wine, beer, buttermilk and the fermentation of bread dough the first controlled biotechnological processes.

Lactobacillus Rye Flour Ferment/Filtrate is the result of such a process – although the biotechnological process used now has nothing to do with the processes as they were carried out thousands of years ago. That is to say, in order to obtain this multivalent resource, rye flour with a high hull content is subjected to a patented biotechnological fermentation process with defined lactobacilli strains (1, 2, 3, 10).

![Fig. 1](image)

Fig. 1 is a schematic depiction of the process and the growth curve for the production of Lactobacillus Rye Flour Ferment/Filtrate. The Lactobacillus Rye Flour Ferment/Filtrate extraction process begins by selectively adding starter cultures to the uniform rye/water mixture (Fig. 1). Following an initial phase, the microorganisms shift to an exponential growth phase, with the population increasing from 10⁷ CFU/ml to 10⁸ CFU/ml during this exponential phase. During the steady-state phase, the metabolic processes of the lactobacilli operate at a rapid rate, which is accompanied by a decrease in pH from pH = 6.5 to pH = 3.5. During this phase, the rye substrate is completely converted to biomass and organic acids. The only groups of compounds that remain unaffected by this process are cellulose and lignans. At the peak of lactobacilli metabolic activity and just before most of the usable substrate is broken down or converted, the process is purposely stopped via pasteurization before the terminal phase is reached.

The result is a sustainable multivalent active ingredient that, especially when applied topically via cosmetics, has a very large range of effective uses (3, 10).

### Composition

The broad range of effective uses is due to the multifaceted nature of this complex active ingredient. Fig. 2 shows the amino acid composition of the Lactobacillus Rye Flour Ferment and Lactobacillus Rye Flour Ferment Filtrate active ingredients.

The materials supply the skin with 6 of the 8 essential amino acids as free amino acids, which is of great importance to the metabolic activity of the living epidermis. In addition, free amino acids are the most significant water-soluble

![Fig. 2](image)

Fig. 2 Composition of the free amino acids formed during the process (10).

<table>
<thead>
<tr>
<th>Research parameter</th>
<th>Analysis value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tocopherol content</td>
<td>1.4 µg/100 ml</td>
</tr>
<tr>
<td>α-tocopherol</td>
<td>1.4 µg/100 ml</td>
</tr>
<tr>
<td>β-tocopherol</td>
<td>&lt; 1 µg/100 ml</td>
</tr>
<tr>
<td>γ-tocopherol</td>
<td>&lt; 1 µg/100 ml</td>
</tr>
<tr>
<td>δ-tocopherol</td>
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<tr>
<td>α-tocopherol</td>
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<tr>
<td>Vitamin B1 content</td>
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<td>Vitamin B2 content</td>
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<tr>
<td>Vitamin B3 content</td>
<td>24 µg/100 ml</td>
</tr>
<tr>
<td>Folic acid content</td>
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<tr>
<td>Pantothenic acid content</td>
<td>83 µg/100 ml</td>
</tr>
<tr>
<td>Nicotinamide content</td>
<td>172 µg/100 ml</td>
</tr>
</tbody>
</table>

Table 1 Vitamin content of the Lactobacillus Rye Flour Ferment Filtrate active ingredient complex (10).
component of the NMF (Natural Moisturizing Factor), essential in maintaining the integrity of the skin barrier. Not only do amino acids play an important role in the retention of moisture in the stratum corneum, they are also the first defense against free radicals.

What is of particular importance is the high proportion of the two acidic amino acids, glutamic acid and aspartic acid, whose high biological relevance to the skin has been the object of research for several years (4).

With regard to vitamins, the high concentrations of pantothenic acid (vitamin B5) and niacinamide are of particular significance. (Table 1)

Pantothenic acid is now commonly used in topical skincare products to soothe the skin. Pantothenic acid is an important component of coenzyme-A and plays an essential role in the energy metabolism of living cells (lipid synthesis). In addition, pantothenic acid stimulates the mitotic activity of cells, leading to the faster regeneration of lesions (7).

Currently, one of the most talked about active ingredients in cosmetics research is niacinamide (niacin, vitamin B3), since pellagra, which is induced by niacin deficiency, is associated with dermatitis. Biochemically, niacin plays an important role in energy metabolism as a component of the cofactors NAD+ and NADP+. In addition, however, it has recently been shown that this material can also make a major contribution to the topical treatment of acne by regulating the production of sebum. Recent studies have shown that, as a cosmetic active ingredient, niacin can stimulate ceramide and cholesterol synthesis in the stratum corneum, thereby making a significant contribution to the stabiliza-
tion of the barrier function (reduction of TEWL). Other in vivo studies show that fine lines and hyperpigmentation are reduced when it is applied topically to mature skin (5, 6).

Besides these groups of substances, other vitamins, polysaccharides (hemicellulose, lignans, cellulose, organic acids, etc.) as well as minerals and trace elements also play an important role in the wide range of effective uses associated with this multivalent active ingredient.

Studies

The high degree of effectiveness of this family of active ingredients was established as early as 2002 in innumerable internal and voluntary application tests by the Wolf family. Fig. 3 shows the normalizing effect of Lactobacillus Rye Flour Ferment through once-daily topical application on a 14-year-old boy (family member) with a clearly visible tendency to skin blemishing. The photos show significant improvement in skin condition after 21 days.

Fig. 4 shows the results of an in-vitro study of the Lactobacillus Rye Flour Ferment Filtrate active ingredient complex on special test bacteria associated with acne, dandruff and atopic eczema (Propionibacterium acnes DSM 1897, Staph. epidermis DSM 20044, Malassezia furfur DSM). Fig. 4 shows the anti-microbial effect of this active ingredient complex in different concentrations on the specified test bacteria.

Even low concentrations of Lactobacillus Rye Flour Ferment Filtrate show significant anti-microbial activity against these three pathogens. Of particular interest is its anti-microbial effectiveness, irrespective of whether a gram + prokaryote or a eukaryotic cell is involved. Such a significant anti-microbial effect cannot be related solely to a decrease in pH value. The literature shows that there is a relationship between probiotics (lactobacilli) and the synthesis of defensin in human epithelial tissue, but the extent to which specific lactobacilli form their own defense system against foreign bacteria has not yet been clarified (8, 9, 10).
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**In vivo verification**

To verify the in vitro data and the personal in vivo study on test subjects with acne vulgaris was carried out to determine, under dermatological supervision, the number of lesions and comedones. The test method used was a single-blind test. In this study, a 2mm layer of active ingredient Woresana Concentrate (rinse-off mask) was applied to one side of the face for a period of 10-15 minutes every 2 days.

Fig. 5 shows the percentage decrease in the number of comedones compared to the number of comedones in the untreated area.

After 6 weeks of treatment, a -73.3 % decrease in the number of comedones compared to the untreated area was observed.

Fig. 6 shows the percentage decrease in the number of lesions using the same treatment regimen (rinse-off mask) compared to the initial situation (Day 0).

After 6 weeks there was a -35.2 % decrease in the number of lesions in the treated area compared to the initial situation (Day 0). In addition to the effect with respect to purely inflammatory parameters, the effect on skin moisture levels (Corneometer CM825, Courage + Khazaka, Germany) and skin roughness (skin microtopography, silicone replica) for the same method of application/treatment was also examined, in each case one day after application. Unlike many commercially-available preparations in this segment, skin moisture levels increased by 6 % on average (in comparison to untreated areas and Day 0) and skin roughness decreased by 7.5 % (in comparison to untreated areas and Day 0). This means that Lactobacillus Rye Flour Ferment not only reduces inflammatory parameters in acne-prone skin, but that the skin's overall health is improved. With regular application, Lactobacillus Rye Flour Ferment active ingredients therefore promote sustained improvement in the condition of the skin [10].

**Summary and Outlook**

The studies and data presented here, in particular the in vivo results, are currently of extreme significance and show that Lactobacillus Rye Flour Ferment products can be incorporated in many ways in topical forms of application. This results in highly-effective skincare products that perfectly combine effectiveness and sustainability. Further in vitro studies on cell cultures and further in vivo verifications in the coming months will better explain the functional mechanisms involved in the efficacy of this product. In addition, other fermentation products using different sustainable grains, together with new lactobacilli species, are currently being developed that will serve to expand the range of products with new potential applications.

It is of particular interest, that Lactobacillus Rye Flour Ferment products are exceptionally well-tolerated by the skin despite the high complexity of their active ingredient, as has been proven in countless studies (human patch tests, Hetcam test, cytotoxicity), and can easily be added into the formulation at the end of any manufacturing process. Not only do these multivalent active ingredient complexes present us with new and sustainable means for providing modern skincare, they also offer proof that humans are complex organisms who have lived in symbiosis with a variety of microorganisms since our species began and that we are, therefore, just another part of nature.
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