KOFA® GRAIN -pH 5-




- Preservative
  for high-moisture grain and grain maize
- Silage additive
  to improve aerobic stability of silage from whole-crop maize and cereals, heavily wilted grass, maize ear products, high-moisture grains, sugar beet pulp, brewer´s grains and distiller´s grains

Problem

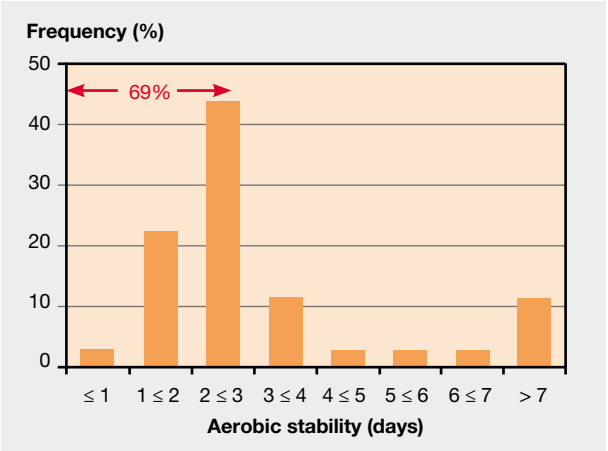
I. High-moisture grain

Freshly harvested grain having moisture content in excess of 14% cannot be stored safely. Massive development of micro-organisms, particularly moulds and yeasts, will spoil the grain, resulting in the breakdown of valuable nutrients and causing severe heating. The moister the grain at harvest, the faster it will deteriorate. The spoilage process accelerates as temperature rises and water is released by microbial metabolic activity. Forced air circulation through the grain stack can stop this acceleration but not the spoilage process *per se*. It can only delay it. Microbial activity in the grain can adversely affect its quality to a greater or lesser degree. In the worst case, grain becomes completely unfit for feeding purposes. There is also a risk of the formation of mycotoxins by storage moulds such as species of the genera *Penicillium* and *Aspergillus*. These fungal metabolites can be extremely dangerous to the health and performance of farm animals.

Compared with drying, preservation of high-moisture grain with propionic acid has become increasingly popular due to cost reasons. This technology has proved to be effective under practical conditions. However, the highly corrosive and caustic properties of this acid and the pungent vapours generated in this process are dangerous for man and machinery. Partial buffering of propionic acid with ammonium and sodium salts, as is done in the production of so-called NC (non-corrosive) products, can produce a gradual improvement of the handling properties but also has a significantly negative impact on its efficacy.

II. Aerobically unstable silages

Deterioration caused by fungi is not only a problem in aerobic storage of high-moisture grain and grain maize but also in silages from various forage crops. Particularly forage maize and maize ear products (CCM and high-moisture maize kernels) tend to spoil when exposed to air after the silo is opened. The following graph exemplarily shows data of an Australian survey (New South Wales Department of Agriculture), where the majority of maize silages rapidly deteriorated upon exposure to air (maximum aerobic stability of 3 days).



Spoilage usually starts with rapid development of yeasts as soon as the silage comes into contact with air. As lactic acid, sugar and other nutrients are broken down into carbon dioxide and water, energy is released, which in turn results in heat generation and temperature rise. Utilization of lactic acid by fungal microorganisms causes an increase in pH, thereby creating perfect environmental conditions for undesired bacteria to thrive. The spoilage process will then continue, accelerated by the higher temperature. Extended breakdown of nutrients, growth of fungi, multiplication of harmful bacteria (enterobacteria, clostridia, listeria) and possibly also the formation of toxic fungal metabolic products (mycotoxins) significantly affect the quality of the silage. It may ultimately lead to total loss of silages for feeding purposes.

The risk of aerobic instability of silages is increased enormously when, due to the high capacity of the harvesting machines and the resulting faster supply of the crop to be ensiled, it is not possible to sufficiently compact forage crop. This will result in a highly porous feed stack, enabling the carbon dioxide produced during fermentation to flow off more easily and oxygen to penetrate more deeply after

opening the silo. The risk of silage heating upon exposure to air is further increased if the silage is removed at too a low feed-out rate which is associated with an unfavourable relation between daily silage requirement for feeding and size of cutting surface of the silo.

The same risk of aerobic spoilage during storage and feeding also frequently occurs in silages produced from industrial by-products such as sugar beet pulp, brewer´s grains and destiller´s grains.

Removal of spoiled top layers can and should be done while unloading, but it requires extra time and labour. However, appearance and smell of the silage do not allways give a reliable information on how deep silage deterioration processes have allready progressed into the silo. Even more important is that instinctive protective reactions of animals, which prevent them from taking in spoilt silage fractions, are no longer effective now as most farmers use total mixed rations for feeding.

Concept

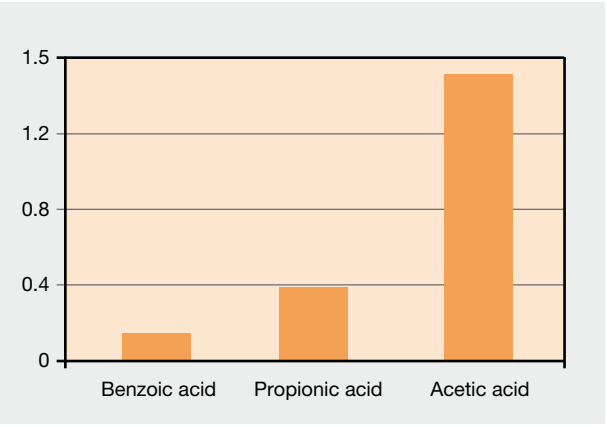
The aim of inventing **KOFA GRAIN -pH 5-** was to formulate a preservative for high-moisture grain which would have an effect similar to that to pure propionic acid, but would offer benefits to the user in terms of safer and easier handling.

Simultaneously, it was intended to make the product suitable for use as a silage additive to prevent quality problems caused by yeasts and moulds, thereby giving the farmer maximum flexibility in controlling similar harmful organisms in different kinds of on-farm produced feed materials and industrial by-products with just one product.

Following an extensive long-term research programme conducted in collaboration with the Oskar-Kellner-Institut für Tierernährung (Institute for Animal Nutrition) in Rostock and the Federal Research Centre of Agriculture (FAL) in Braunschweig, an effective combination of preservatives has been developed in a user-friendly formulation.

The choice of preservatives was based on their inhibitory effect on moulds and yeasts. Besides propionic acid as the basic component, **KOFA GRAIN -pH 5-** also contains the active ingredient sodium benzoate. Sodium benzoate is the salt of benzoic acid, which is much more effective than propionic acid in suppressing harmful fungi. The results obtained by FAL Braunschweig are shown in the graph below. They demonstrate the effect of various organic acids on the most frequent storage mould in silages, *Penicillium roqueforti*. The graph clearly shows the much stronger effect of benzoic acid, which is released from its salt in weakly acidic conditions.

Required concentration to suppress growth of *Penicillium roqueforti* (%)



KOFA® GRAIN -pH 5-

Product

KOFA GRAIN -pH 5- is a stable, aqueous solution which contains maximum concentrations of each of the following ingredients:

- propionic acid,
- sodium benzoate and
- sodium propionate.

The reddish coloured liquid has a slightly pungent smell of propionic acid; it is slightly acidic (pH approx. 5.0) and has a density of about 1.1 g/cm^3.

KOFA GRAIN -pH 5- is a ready-to-use product. It must not be mixed with water. The dosing equipment must not be rinsed with water, as this can cause precipitation of crystalline benzoic acid.

The beneficial antimicrobial properties of **KOFA GRAIN -pH 5-** in high-moisture grain and high-moisture maize originate from the combined effect of the preservatives used. The sum of the individual effects of each components applied at equivalent dosage will not produce the same result.

The propionic acid contained in the product is partly buffered by sodium benzoate and sodium propionate, so it is significantly less aggressive and less corrosive. Alongside this, the fraction of free propionic acid still remaining in the mixture activates the antimycotic potential of sodium benzoate. This is the decisive pre-requirement for the positive effect of **KOFA GRAIN -pH 5-** in non-fermenting feedstuffs, e.g. high-moisture grains.

Use and dosage

I. High-moisture grain

The minimum application rate of **KOFA GRAIN -pH 5-** for preservation of high-moisture grain and high-moisture maize depends on moisture levels of the material at harvest and intended storage length (Table 1).

Table 1:
Dosage of KOFA GRAIN -pH 5- for preservation of high-moisture grain and high-moisture maize

Grain type / moisture content	Minimum dosage (litres/t) Intended storage length (months)		
WHEAT, BARLEY, OATS, RYE, TRITICALE, MAIZE	to 3	3 to 6	6 to 12
14 %	3.0	3.5	4.0
16 %	4.5	5.0	5.5
18 %	5.0	6.0	7.0
20 %	6.0	7.0	8.0
22 %	7.0	8.0	9.0
24 %	8.0	9.0	10.0
26 %	9.0	10.0	11.0
28 %	10.0	11.0	12.5
30 %	11.0	12.5	14,5
>30 to 35 %	- -	14.5	17.0
>35 to 40 %	- -	16.5	18.5
>40 to 45 %	- -	18.5	20.0

Application rates apply to freshly harvested grain which contains only little contamination (chaff, weeds, dust) and which has not been previously damaged by intermediate storage.

Safety margins of 10 -15% need to be added to minimum dosages for each of the following conditions:

- Treatment of grain containing a high leval of contamination
- Treatment and storage at high temperatures (> 30°C)
- Treated grain conveyed pneumatically
- Storage of processed (milled, crimped) grain upon unrestricted exposure to air.

II. Silages

When using **KOFA GRAIN -pH 5-** as a silage additive, the following typical application rates apply:

3 to 5 litres per tonne of crop.

Application rates vary depending on forage type and expected risk of aerobic spoilage. The application rates listed in Table 2 are recommended for the production of silages with **KOFA GRAIN -pH 5-**.

Table 2:
Application rates of KOFA GRAIN -pH 5- as silage additive

Crop	DM content (%)	**KOFA GRAIN -pH 5-** (l / t)
Treatment of entire feed stack		
Forage maize	28 - 35	3.5
	35 - 40	4.0
	> 40	4.5
Whole-crop cereals	< 35	3.5
	35 - 50	4.0
	> 50	4.5
Heavily wilted grass	40 - 50	3.0
	50 - 60	4.0
	60 - 70	5.0
Maize ear products (CCM, high-moisture maize kernels)	55 - 65	3.0
	65 - 70	4.0
	70 - 75	5.0
Processed (milled, crimped) high-moisture grain	55 - 65	3.0
	65 - 70	4.0
	70 - 75	5.0
Brewer´s grains	25 - 35	4 - 5
Destiller´s grains	20 - 30	4 - 5
Sugar beet pulp		
melassed	20 - 28	3 - 4
non-melassed	20 - 26	4 - 5
Partial treatment of the feed stack (upper third part)		
Forage maize	28 - 35	4 - 5
Whole-crop cereals	35 - 60	4 - 5
Heavily wilted grass	40 - 70	4 - 5

Application

In order for **KOFA GRAIN -pH 5-** to show its full effects, it must be homogeneously mixed into the material to be treated. This is ideally achieved by applying the product when the crop is harvested or the feed material produced, i.e. in an auger for high-moisture grain and grain maize, on the forage harvester, self-loading forage wagon or baling press for silage crops (forage maize, grass, whole-crop cereals), in the mill or crimper for processed grain products to be ensiled (CCM, high-moisture maize kernels, high-moisture grain) or, for industrial by-products (brewer's grains, distiller´s grains and sugar beet pulp), in the plant in which they are produced. **KOFA GRAIN -pH 5-** is applied using any commercially available dosing equipment for liquid products. We recommend applicators supplied by SILA GmbH, Bitterfeld (marketed under the brand name SILASPRAY).

Tests of KOFA GRAIN -pH 5- as a preservative

Intensive scientific research was performed on the efficacy of **KOFA GRAIN -pH 5-** as a preservative for high-moisture grain and grain maize over several years at the Oskar-Kellner-Institute in Rostock and at the Institute for Grassland and Forage Research at the Federal Research Centre of Agriculture (FAL) in Braunschweig. These tests were carried out on different grain species with different moisture contents at harvest.

The preserving effect of **KOFA GRAIN -pH 5-** was evaluated using the parameter infection rate of storage moulds. This parameter relates to the proportion of individual grain kernels which are affected by moulds growth.

In the preservation of wheat with 21% moisture content, did the addition of 5 l/t of **KOFA GRAIN -pH 5-** reliably prevent mould development over a period of at least 10 weeks. The single use of sodium benzoate alone did not result in this effect, even at a high dosage (Table 3).

Another trial using wheat with the same moisture content compared the effects of **KOFA GRAIN -pH 5-** and propionic acid applied at the same rate (5 l/t) over a period of 29 weeks (Table 4). Both additives successfully prevented mould growth. Taking into consideration the quantity of the total active substance applied, however, **KOFA GRAIN -pH 5-** was clearly more effective (3,400 ppm total active ingredients, thereof 2,000 ppm propionic acid) than pure propionic acid (5,000 ppm propionic acid).

The use of **KOFA GRAIN -pH 5-** also markedly inhibited deterioration of high-moisture grains over an even longer storage period (8 months) (Table 5).

Table 3: Effect of KOFA GRAIN -pH 5- on infestation of grain by storage moulds

Treatment / Dosage	Storage fungi infection rate (%) when stored for	
	6 weeks	10 weeks
Control	82	100
Sodium benzoate solution 10 l / t	100	100
KOFA GRAIN -pH 5- 2 l / t 5 l / t 10 l / t	 91 0 0	 97 0 1

Oskar-Kellner-Institut, Rostock

Table 4: Effect of KOFA GRAIN -pH 5- and propionic acid on infestation of grain by storage moulds

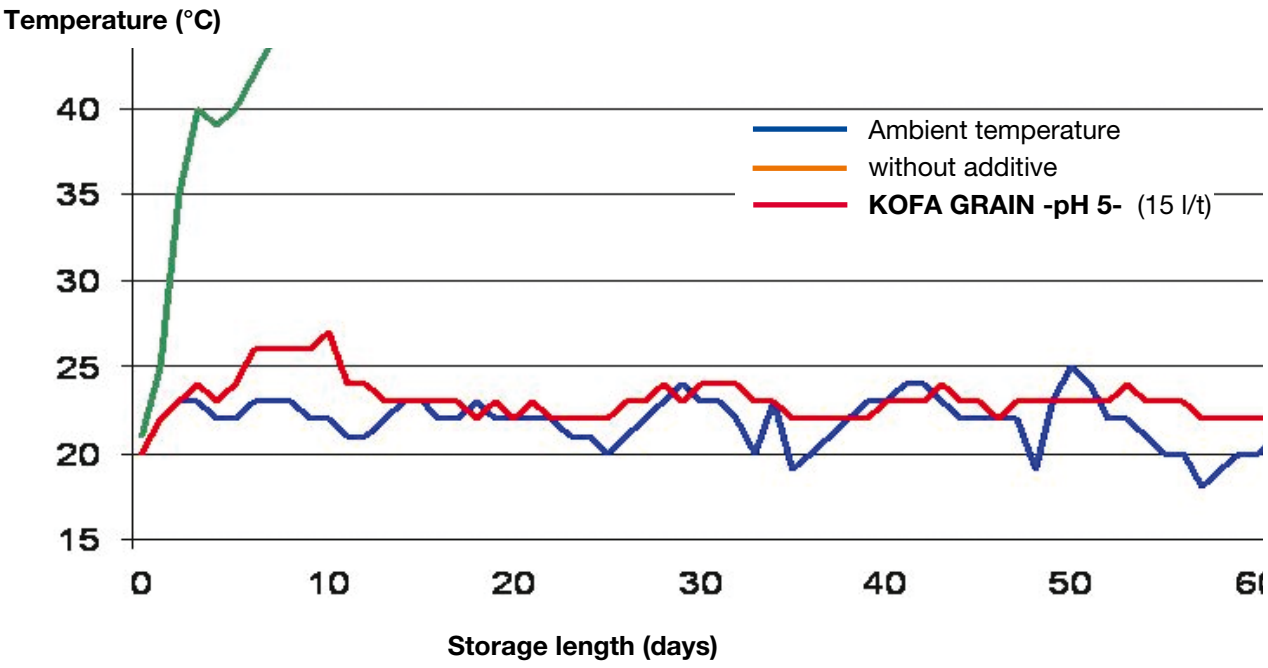
Treatment	Storage fungi infection rate (%) when stored for ... weeks			
	4	8	12	29
Control	100	100	100	n.d.
Propionic acid (5 l / t)	1	0	0	0
KOFA GRAIN -pH 5- (5 l / t)	0	0	0	0

Oskar-Kellner-Institut, Rostock; n.d. = not determined

Table 5: Effect of KOFA GRAIN -pH 5- on infestation of grains by storage moulds

Trial (%)	Moisture content (l/t)	**KOFA GRAIN -pH 5-** (%)	Storage fungi infection rate
Barley	27	0 9	100 1
Barley	20	0 7	96 0
Wheat	20	0 7	61 0

The results given in the graph below clearly illustrate that **KOFA GRAIN -pH 5-** can also be used successfully as a preservative for whole maize kernels with high moisture content (about 35%) at harvest. The untreated high-moisture maize deteriorated rapidly as reflected by significant temperature increase after a very short storage period. This was caused by metabolic activity of micro-organisms, such as moulds and yeasts. When **KOFA GRAIN -pH 5-** was used, temperature remained stable over the entire test period. Growth and metabolic activity of harmful fungi were suppressed.



The efficacy of **KOFA GRAIN -pH 5-** was also confirmed in the preservation of high-moisture grain and grain maize under practical conditions. To test the effects of **KOFA GRAIN -pH 5-** on mycological quality and mycotoxin formation during storage, controlled studies were performed in 2003 and 2004 in collaboration with the Agricultural Analysis and Research Institute (LUFA) Nordwest (Oldenburg) and LUFA Rostock, in which fungal counts and level of ochratoxin A, the main mycotoxin of storage moulds, were determined.

For this purpose,
a total of 43 batches of grain and maize, including
- 11 batches of barley,
- 8 batches of wheat,
- 8 batches of triticale and
- 16 batches of maize

with differing initial moisture content at harvest were treated with **KOFA GRAIN -pH 5-** at the recommended application rates on 26 selected farms in Germany. A total of about 2,300 tonnes of grain (barley, wheat, triticale) and about 1,600 tonnes of maize grain was preserved, and subsequently quality of the stored crop was monitored. Fungal counts and ochra- toxin A content were determined at regular intervals (approximately every 3 months) over a period of up to 15 months.

Without exception, all batches proved to have been very well preserved. Both, fungal count for product-typical moulds and spoilage-indicating moulds confirmed the superior mycological quality of all 137 samples examined. Yeasts were only rarely detected and never exceeded a value of 10,000 CFU/g, and as such were negligible.

The analysis for ochratoxin A in 123 samples only produced a positive result in two cases (1.6% of the total sample pool). However, the values were always in the range of the detection limit and were therefore toxicologically irrelevant.

Tests of KOFA GRAIN -pH 5- as a silage additive

The efficacy of **KOFA GRAIN -pH 5-** to improve bunk-life of silage exposed to air was demonstrated repeatedly in a number of tests at several different international and independent research facilities (Table 6).

Table 6: Effect of KOFA GRAIN -pH 5- on aerobic stability of silage

Research Facility	Forage type	DM content (%)	Dosage (l / t)	Aerobic stability (days)	
				without additive	**KOFA GRAIN -pH 5-**
A	Forage maize	30	5.0	2.7	9.0
B	Forage maize	39	5.0	1.4	3.4
	Grass	48	5.0	1.3	6.4
C	Grass	44	3.0	1.0	5.2
	Grass	44	4.0	1.0	4.3
D	Grass-clover mix	30	4.5	1.2	6.7
	Clover grass	60	4.5	3.3	8.0
	Whole crop barley				
	10 cm chop length	26	5.0	1.0	6.0
	2 cm chop length	26	5.0	1.3	7.0
	Whole crop barley				
	10 cm chop length	33	5.0	4.0	7.0
	2 cm chop length	33	5.0	3.7	7.0
E	Sugar beet pulp				
	unmolassed	23	4.0	1.0	6.6
	molassed	25	4.0	5.0	7.0

A - LWK Chamber of Agriculture Rhineland; **B** - Brandenburg Regional Agricultural Institute, Paulinenaue;
C - LWK Chamber of Agriculture Northrhine-Westfalia; **D** - Swedish Agricultural University, Uppsala; **E** - Humboldt University, Berlin

KOFA® GRAIN -pH 5-

As ensiling of processed (crimped) grains has attracted significant attention also in regions other than Scandinavia or Great Britain, where it is a well-established technology, studies have been carried out on the use of **KOFA GRAIN -pH 5-** in this application by several research bodies, e.g. the University of Bonn, Germany and the Agricultural Research Centre of the State Saxony, Köllitsch, Germany.

Data summarized in Table 7 clearly show that **KOFA GRAIN -pH 5-** markedly enhances aerobic stability of crimped grains which had been stored anaerobically prior to opening the silo. This effect did neither depend on grain moisture content at harvest nor on grain type.

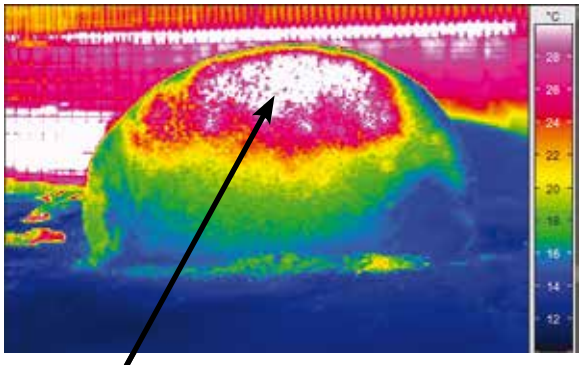
Table 7: Effect of KOFA GRAIN -pH 5- on aerobic stability of crimped and ensiled grains

Research Facility	Forage type	DM content (%)	Dosage (l / t)	Aerobic stability (days)	
				without additive	**KOFA GRAIN -pH 5-**
A	Maize, crimped	65	3.0	4.0	14.0
B	Maize, crimped	62	3.0	2.0	> 10.0
A	Barley, crimped	83	2.0	10.0	10.0
			4.0	10.0	10.0
A	Barley, crimped	76	2.0	1.2	10.0
			4.0	1.2	10.0
A	Barley, crimped	70	4.0	1.5	10.0
A	Barley, crimped	69	2.0	1.6	2.2
			4.0	1.6	7.4
A	Wheat, crimped	82	2.0	10.0	10.0
			4.0	10.0	10.0
	Wheat, crimped	79	2.0	1.0	3.3
			4.0	1.0	3.1
A	Wheat, crimped	73	4.0	1.7	10.0
A	Wheat, crimped	70	2.0	1.4	8.6
			4.0	1.4	10.0

A - University of Bonn, Germany **B** - Agricultural Research Centre of the State Saxony, Köllitsch, Germany

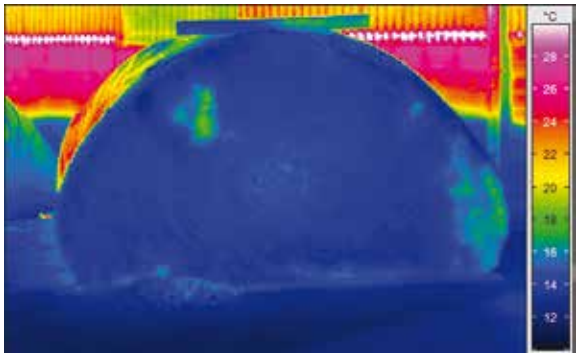
Further experiments were performed by the University of Bonn in 2005 and 2006. They showed excellent effects of **KOFA GRAIN -pH 5-** on aerobic stability of crimped grains which were ensiled in plastic tubes (company Ag-Bag, Germany). In all trials were 4 l/t of **KOFA GRAIN -pH 5-** sufficient to significantly improve aerobic stability (see exemplarily thermopictures of crimped barley, 31 % moisture).

Control (no additive)



> 40 °C strong heating

KOFA GRAIN -pH 5- (4 l/t FM)



no heating

KOFA® GRAIN -pH 5-

Patent

The formula, the production and the use of **KOFA GRAIN -pH 5-** as a preservative for non-fermenting feedstuffs, particularly in high-moisture grain and grain maize, is protected by international patents (European patent no. EP 0 803 200 B1; US patent no. 5,906,849; Canadian patent no. 2,203,344).

Animal feed additive legislation in European Community

KOFA GRAIN -pH 5- contains active substances whose properties and harmlessness have been proven for quite some time, and which are also frequently used as preservatives in foods and beverages. **KOFA GRAIN -pH 5-** has been approved in the EC without a time limit as technological additive (category 1), preservatives (functional group a) for preservation of high-moisture grain, under REGULATIONS (EC) no. 1876/2006 of 18 December 2006 and 757/2007 of 29 June 2007. **KOFA GRAIN -pH 5-** carries the official identification number E 700.

Approval by German Agricultural Society (DLG)

Based on efficacy trials by independent research facilities according to official guidelines, the German Agricultural Society (DLG) has awarded the preparation **KOFA GRAIN -pH 5-** the

DLG seal for silage additives for aim-of-action 2, "Improving aerobic stability".

In conclusion, **KOFA GRAIN -pH 5-** is a highly effective, comprehensively tested and user-friendly product

- for inhibiting mould growth and mycotoxin formation in aerobically stored high-moisture grain and grain maize and
- for preventing aerobic deterioration and heating as well as mycotoxin formation in silages after silo opening.

The use of **KOFA GRAIN -pH 5-** maintains the nutritional value and hygienic quality of the feed. The product offers the user maximum flexibility, as it can be used to solve similar problems in different farm-derived feedstuffs and industrial by-products.

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