Feed Additives, can they improve animal welfare?

J. Brufau, R. Lizardo and B. Vilà.
IRTA

27/8/2014, Copenhagen
Welcome to our world

Now that scientists have belatedly declared that mammals, birds and many other animals are conscious, it is time for society to act, says Marc Bekoff.

ARE animals conscious? This question has a long and venerable history. Charles Darwin asked it when pondering the evolution of consciousness. His ideas about evolutionary continuity—that differences between species are differences in degree rather than kind—lead to a firm conclusion: “they” (other animals) have it too.

In July of this year, the question was discussed in detail by a group of scientists gathered at the University of Cambridge for the first annual Francis Crick Memorial Conference. Crick, discoverer of DNA, spent the latter part of his career studying consciousness and in 1994 published a book about it, The Anishing Hypothesis: The Scientific Search for the Soul. The upshot of the meeting was the Cambridge Declaration on consciousness, which was publicly proclaimed by three recent neuroscientists, David Israel of the Neurosciences Institute in La Jolla, California, and Low of Stanford University, and Toshif Koch of the California Institute of Technology.

The declaration concludes that human animals have the anatomical, neurochemical, other creatures, including octopuses, also possess these neurological substrates. My first take on the declaration was incredulity. Did we really not need this statement of the obvious? Many renowned researchers reached the same conclusion years ago. The declaration did not include fish, because the evidence supporting consciousness in this group of vertebrates is also compelling.

Nevertheless, we should applaud them for doing this. The declaration is not aimed at the US. I’m concerned that those who ignore science. The Treaty of Lisbon into force...
The objective of the presentation is:

- Improvement of performances through gut health is AW indicator?
- Is the health improvement easily measurable?
- What are the main indicators to be considered?
- Can these indicators be connected to animal performance?
- How is gut immunity involved in animal performance?
What does animal welfare mean in a regular farm?

- Stress induces a General Adaptation Syndrome (Selye 1950).

- Stress affects the hormonal control of metabolism, reproduction, growth and immunity.

- Conclusion: the animal adaptive response to stress is the integration of multiple, often interactive, hormone responses that directly affect health and well-being.
General Adaptation Syndrome, Selye 1950

Biological sense **STRESS** is the interaction between **damage** and **defense**
Naturally, farm animals are challenged by different stressors

- **“All farm animals** will experience **some level of stress** during their lives. Stress reduces the fitness of an animal, which can be simply expressed through failure to achieve production performance standards or targets, or more drastically, through disease and death” *(Mario Rostagno 2009)*.

- **Stress factors which affect animal production**:
  1. Inadequate nutrition
  2. Deprivation of water/ or feed
  3. Heat/Cold
  4. Overcrowding
  5. Handling (interaction human manipulation)
“Stress and the Gastrointestinal Tract”

- The enteric nervous system (ENS) is an integrated network located within the wall of the gastrointestinal tract. (Brain-Gut interaction).

- **Stress** may not only be responsible for functional disorders, but may contribute to inflammatory disorders and infections of the gastrointestinal tract.

- Neurotransmitters play a role in animal responses to challenges/stressors (Noreadrenaline-naturally intestinal mucosal).

- There is crosstalk between neuroendocrine and immune systems.

- An imbalance on these systems in response to stress can lead to significant changes in immune response and consequently susceptibility to infection.
Schematic representation of intestinal anti-inflammatory reflex (Niewold 2014)

Figure 1 - Schematic representation of the intestinal anti-inflammatory reflex. Feed compounds can give a pro-inflammatory (+) stimulus to enterocytes and macrophages. This leads to the production of pro-inflammatory interleukins (ILs), which also reach the brain. A down (-) regulatory signal is returned to the intestine through the nervus (N) vagus. Adapted after Niewold, 2013.

II. BIOMARKERS FOR GUT HEALTH IN POULTRY
Effect of noradrenaline on the growth of Campylobacter

Figure 2. Effect of noradrenaline (100 μM) on the growth of C. jejuni in iron-restricted media (DMEM containing 10% serum). Closed circles show the growth profile of the control cultures. Open triangles show cultures plus noradrenaline. Data from Thomas et al. (unpublished).
Stress and the Gastrointestinal Tract

Stress releases catecholamine and this results in:

I. Decreased gastric acid production
II. Delayed gastric emptying
III. Accelerated intestinal motility
IV. Accelerated colonic transit

Consequently increased pH in the stomach increases probability of survival of food borne pathogens (E. coli, salmonella and Campylobacter) and colonization of the gastrointestinal tract.
Feed intake / Neuroendocrine control of appetite during the stress response

Feed intake is necessary for the growth and survival of all animals, it is important for us to understand how common stressors reduce feed intake at the biochemical level, with the hope of someday being able to prevent or diminish appetite loss and subsequent reduction in the growth, health and well-being of animals.
New European model of animal production since 2002

- Animal Production should be sustainable in the EU and based on:
  - Animal Protection
  - Consumer Protection
  - Environment protection

Travelling to 2030; via S.E.T
Feed additives

- Regulated By EC 1831/2003

- Substances, micro-organisms or preparations, other than feed material and premixtures, which are intentionally added to feed or water in order to perform, in particular, one or more of the functions mentioned in Article 5(3)

  ✓ Favourably affect the characteristics of feed or animal products
  ✓ Favourably affect the colour of ornamental fish and birds
  ✓ Satisfy the nutritional needs of animals
  ✓ Favourably affect animal production, performance or welfare
  ✓ Have a coccidiostat or histomonostatic effect
Outline questions

- Why Animal Welfare criteria are not yet implemented in Feed Additive evaluation?

  The concept of Animal Welfare is under revision in EU. Strategies are in progress 2012-2015.

- Which parameters are much more accepted by farmers in order to consider Animal Welfare benefits?

- Feed additives, may they play a role on animal welfare assessment?

- Feed additive have to be evaluated under **Good Health** conditions?
A zootechnical additive is any additive used to favourably affect the performance of animals in good health, or to favourably affect the environment.
The purpose was to:
- examine the scientific basis for the existing functional groups
- propose, if necessary, based on this review, the establishment of additional functional groups (or categories).

Potential new categories
1. Additives which favorably affect animal welfare:
   Metabolic regulators, Immuno-modulators, Detoxifiers.

2. Additives which improve product quality:
   Microbial contamination controllers, Nutritional value enhancers, Sensory additives.
How to improve AW at the farm level

1.- Improve management of animals.

2.- Better knowledge of Feeding programs and feed composition.

3.- Supplementation of diets with alternative additives to AGP.
Enriched Cages for laying hens
Pig production 2030

EuroTier 2012

Big Dutchman
Gestation sows in free stalls
ABSTRACTS
OF
LECTURES
AND
POSTERS

The International
Debate Conference
for the Feed & Food Chain

Antimicrobial Growth Promoters:
Worldwide Ban on the Horizon?

31 January-1 February 2005

Grand Hotel Huis ter Duin, Noordwijk aan Zee, the Netherlands
Assessment of alternatives substances
Animal nutrition and Gut microflora interactions (Animal protection)

- Growth promoting agents
- Improved sanitation/husbandry
- Reduced antigenic load
- Increased nutrient bioavailability (?)
- Improved health status
- Nutrient sparing
- Performance response
- Reduced mortality/morbidity
WAR AND PEACE AT MUCOSAL SURFACES

Philippe J. Sansonetti

Abstract | The gut contains a diverse array of bacteria that can affect the body's immune system, particularly in the gut and effector systems that are available to trigger inflammatory or innate immune responses to microbe invasion. So, a fine line seems to exist between the homoeostatic balance maintained in the presence of commensal gut flora and the necessarily destructive response to bacterial pathogens that invade the gut mucosa. This review discusses the mechanisms for establishing and controlling the "dialogue" between unresponsive and infected immune defences in the gut. S. vis pacem, pax bella. (If you wish for peace, prepare for war.)

Figure 2 | Bacteria trigger a pro-inflammatory programme in intestinal epithelial cells, using various strategies. Pathogenic bacteria and possibly commensal bacteria are detected by epithelial cells through cell-surface receptors (such as Toll-like receptors, TLRs) and by recognition of microbial products. Detection by TLRs or other intracellular pattern recognition receptors triggers a signalling cascade that leads to the activation of nuclear factor κB (NF-κB), which translates to the nucleus, where it promotes the transcription of pro-inflammatory genes. Some pathogenic bacteria such as Citrobacter rodentium and bacillus subtilis activate epithelial cells and secretory cells, which induce NF-κB activation. In contrast, enteropathogenic Escherichia coli (EPEC), enterohemorrhagic E. coli (EHEC), and Helicobacter pylori elicit a pro-inflammatory response in the cell through TLR 4, type I IFN-α/β and secretory IgA systems. Different mechanisms are used by Shigella spp. and Salmonella spp., which directly invade the cell, resulting in NF-κB activity via TLR 4 and thus triggering an inflammatory response. TNF-α, pathogen-associated molecular patterns.

Figure 3 | Expression of TLRs and NODs by luminal surface versus crypt epithelial cells in the small intestine. This scheme shows the probable differences between the epithelial cells at the luminal surface and those in the crypts of the gut in terms of their expression of pattern recognition receptors such as Toll-like receptors (TLRs) and nucleotide-binding oligomerization domain (NOD) proteins for sensing the presence of microorganisms through their pathogen-associated molecular patterns. To protect stem cells and their environment, crypts are organized as a layered units of bacterial sensing and destruction, in which Paneth cells through their production of defensins play an important role. A similar pattern is likely to occur in the colon, in which Paneth cells are absent, and defensins are produced by epithelial cells. CUBIC, CSG, and MUC2 are shown.
Mucosal surfaces place for “dialogue”

The intestinal epithelium: an interactive barrier

.- Physical barrier

.- Innate immunity

.- Adaptive immunity

Crosstalk between commensals and mucosae

Crosstalk between pathogens and mucosae

Philipe J. Sansonetti 2004
Alternative feed additive products

- Organic acids
- Enzyme preparations
- Micro-organisms (Probiotics)
- Oligosaccharides (Prebiotics)
- Immunity enhancers
- Highly available minerals
- Herbs and essential oils
Nutritional and other studies, some examples
Some factors affecting wet litter in commercial poultry flocks

- Leaking drinkers
- Poor ventilation
- High dietary K or Na
- High dietary saturated fat
- Poor litter absorption
- High stocking density
- Microbial enteritidis
- High humidity
- Wet litter

Williams 2005
Williams, 2005
Integrated disease management by maintenance of gut integrity
Figure 2. The intercurrent coccidiosis-NE syndrome: a network of potentially important pathophysiological, medicinal, nutritional and husbandry factors. Those with solid-line arrows and ellipses are beneficial in controlling disease, those with dashed-line arrows and ellipses impart high disease risk. Major high-risk relationships are shown by double-line arrows. AGP, antibiotic growth promoters; CIA, chick infectious anaemia; CEP, competitive exclusion product; Cp, Clostridium perfringens; IBD, infectious bursal disease; MD, Marek's disease; NE, necrotic enteritis.
Effect of xylanase and/or monensin on performance, coccidiosis infection and digesta viscosity of chickens challenged with *Eimeria spp.*

<table>
<thead>
<tr>
<th></th>
<th>BW 29 d</th>
<th>FCR 0-29 d</th>
<th>E. lesions Sum (21d)</th>
<th>Viscosity cps (14d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Not challenged</td>
<td>1457</td>
<td>1.53</td>
<td>0</td>
<td>14.9</td>
</tr>
<tr>
<td>Challenged</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>1400</td>
<td>1.58</td>
<td>6.8</td>
<td>9.8</td>
</tr>
<tr>
<td>Monensin</td>
<td>1443</td>
<td>1.49</td>
<td>3.4</td>
<td>8.9</td>
</tr>
<tr>
<td>Enzyme</td>
<td>1421</td>
<td>1.53</td>
<td>4.3</td>
<td>4.7</td>
</tr>
<tr>
<td>M + E</td>
<td>1513</td>
<td>1.49</td>
<td>3.5</td>
<td>5.6</td>
</tr>
</tbody>
</table>

*Inoculation*  
**Monensin**  
*Enzyme*  
*Interaction*

Francesch *et al.*, 2008
Examples: Efficacy assessment on immune processes

Short communication

β 1-4 mannobiose enhances Salmonella-killing activity and activates innate immune responses in chicken macrophages

Masahisa Ibuki1, Jennifer Kovacs-Nolan2, Rensuke Fukui1, Hiroyuki Kanatani3, Yoshinori Mine4,5

1 R&D Institute, R&D Co., Ltd., 1 Fuchisaka-cho, Sumiyoshi-ku, Osaka 558-8541, Japan
2 Department of Food Science, University of Guelph, Guelph, Ontario, Canada N1G 2W1
3 Tsukuba R&D Center, R&D Co., Ltd, 4-3, Kanamachi, Tsukubakai-cho, Ibaraki, Japan

Fig. 1. Effect of MNB on phagocytic activity of chicken macrophages. MQ-NCSU cells were treated with increasing concentrations of MNB for 2 h, followed by incubation with fluorescein-labeled E. coli BioParticles. Data shown are mean ± SEM. Results are expressed as % phagocytosis relative to untreated cells. *P < 0.05 compared to untreated cells.
**Table 4.** Effects of yeast cell wall (YCW) on the relative lymphoid organ weight\(^1\) and the delayed cutaneous hypersensitivity reaction of chicken\(^2\) inoculated with LPS of *E. coli*

<table>
<thead>
<tr>
<th>Effect</th>
<th>Spleen</th>
<th>Bursa of Fabricius</th>
<th>Delayed cutaneous hypersensitivity reaction 14 d (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>YCW</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0 mg/kg</td>
<td>0.125</td>
<td>0.290</td>
<td>0.301</td>
</tr>
<tr>
<td>500 mg/kg</td>
<td>0.112</td>
<td>0.304</td>
<td>0.441</td>
</tr>
<tr>
<td>LPS-<em>E. coli</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Without challenge</td>
<td>0.117</td>
<td>0.331</td>
<td>0.326</td>
</tr>
<tr>
<td>With challenge</td>
<td>0.120</td>
<td>0.263</td>
<td>0.416</td>
</tr>
<tr>
<td>YCW LPS-<em>E. coli</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No No</td>
<td>0.114</td>
<td>0.348(^a)</td>
<td>0.238(^c)</td>
</tr>
<tr>
<td>Yes No</td>
<td>0.120</td>
<td>0.314(^a)</td>
<td>0.414(^a)</td>
</tr>
<tr>
<td>No Yes</td>
<td>0.136</td>
<td>0.232(^b)</td>
<td>0.365(^b)</td>
</tr>
<tr>
<td>Yes Yes</td>
<td>0.105</td>
<td>0.294(^a)</td>
<td>0.467(^a)</td>
</tr>
<tr>
<td>SE</td>
<td>0.016</td>
<td>0.024</td>
<td>0.050</td>
</tr>
</tbody>
</table>

Source of variation (**P**)

- YCW: 0.42
- LPS-*E. coli*: 0.84
- Interaction: 0.24

---

\( ^1 \)\( n = 11 \) chickens.

\( ^2 \)\( n = 8 \) chickens.

\( ^a, ^b \)Within a column, values not sharing a common superscript letter are significantly different (**P** \( \leq 0.05 \)).
Immune responses to dietary β-glucan in broiler chicks during an *Eimeria* challenge

C. M. Cox,* L. H. Sumners,* S. Kim,* A. P. McElroy,* M. R. Bedford,† and R. A. Dalloul*1

*Avian Immunobiology Laboratory, Department of Animal and Poultry Sciences, Virginia Tech, Blacksburg 24061; and †AB Vista Feed Ingredients, Marlborough, Wiltshire, SN8 4AN, United Kingdom

Figure 1. Effect of β-glucan supplementation on gross intestinal lesions scores of Cobb 500 broiler chicks on d 14 (6 d post *Eimeria* infection). Data are represented as least squares means ± SEM. YGT = Auxoferm YGT, *Saccharomyces cerevisiae*-derived β-glucan. There was a significant effect of dietary treatment in the duodenum (*P = 0.04) and jejunum (**P = 0.02).

2010 Poultry Science 89 :2597–2607
Limited Treatment with $\beta$-1,3/1,6-Glucan Improves Production Values of Broiler Chickens Challenged with *Escherichia coli*

G. R. Huff,* W. E. Huff,* N. C. Rath,* and G. Tellez†

*USDA, Agricultural Research Service, Poultry Production and Product Safety Research, and Department of Poultry Science, University of Arkansas, Fayetteville 72701
β-Galactomannan and *Saccharomyces cerevisiae* modulate Immune response in pigs
B-galactomannan and *Saccharomyces cerevisiae* modulate Immune response in pigs

Badia et al.

**FIG 3** Effects of *S. cerevisiae* var. *bouardii* (Scb) and βGM on cytokine and chemokine mRNA expression in IECs cultured with *Salmonella*. IECs (1 × 10⁶ cells/well) were cocultured with *S. cerevisiae* var. *bouardii* (3 yeast cells/cell) or βGM (10 μg/ml) with *Salmonella* (MOI of 4) for 3 h. Data (n = 6) are presented as means of mRNA relative expression ± SDs. Columns with no common superscripts are significantly different (P < 0.05). Results are representative of 3 independent experiments. □, control; ☐, *Salmonella*. 

27 /8/2014, Copenhagen
Probiotics (Direct feed microbial)

Preliminary update on functionality of probiotics in poultry and pig feeding.

Functionality of probiotics application, review from 1995 until now.
Scientific probiotic studies in monogastric animals published since 1995 until now. Data bases from “Web of Science Core Collection”

**Probiotics / Poultry**
- Total number of references: 474
- Bacillus spp.: 62
- Saccharomyces spp.: 114
- Full article found: 421
  - Bacillus spp.: 54
  - Saccharomyces spp.: 95
  - LAB (Lactococcus / Pediococcus / Bifidobacterium / Enterococcus / Lactobacillus): 34
  - For data extraction: 131

**Probiotics / Pig**
- Total number of references: 399
- Bacillus spp.: 51
- Saccharomyces spp.: 91
- Full article found: 258
  - Bacillus spp.: 47
  - Saccharomyces spp.: 64
  - LAB (Lactococcus / Pediococcus / Bifidobacterium / Enterococcus / Lactobacillus): 22
  - For data extraction: 113
Evolution of main parameters measured in probiotic poultry and swine studies
Example of Targeting microbiota / Bacillus spp

Bacillus spp / swine

Bacillus spp / poultry

- Enhance gut development
- Lower gut pH
- Improve Digestibility
- Boost Immunity
- Control enteric pathogens
- Improve Health status (lower incidence of diarrhoea / lower ammonia emissions)
Suggested **End-points** for demonstration of efficacy on Animal welfare

**In vitro studies**: most of the experiments conducted until now, however they are essential for the first step.

**In vivo studies**: to conduct studies with animals under certain conditions and to assess the benefits of the products on the mucosal and epithelial cells from intestine.

Morphology, Immunity reaction and Microflora development.

i.e. **Blood analysis** - cortisol, heat shock protein, neutrophils/lymphocytes,

i.e. **Mucosal** - epithelial morphology, innate immunity of IEC.

i.e. **Microflora** - Reduction of zoonotic bacteria population.

The animal performance studies may be also involved in order to justify the interaction between AW and performance improvement.
Are we able to answer all the questions generated?

Improvement of performances through gut health is AW indicator?

Is health improvement easily measurable?

What are the main indicators to be considered?

Can this indicators be connected to animal performance?

How is gut immunity involved in animal performance?
Are we able to answer all the questions generated?

The most important action will be to understand the interaction between Animal welfare and the concept of stress and the physiology of the gastrointestinal tract.

Animal health improvement is difficult to assess, especially when we are dealing with benefits of Feed Additives in order to satisfy Animal welfare indicators.

The indicators should be clearly well identified under stress conditions first.

Immune indicators must be considered to determine the degree of animal defense in order to prevent damage by the stressors.
Monogastric Nutrition subprogram