Defining Post-Dairy Digestive Discomfort Beyond Lactose Intolerance

Andrew J Clarke, PhD
After this briefing, the attendees will be able to...

• Discuss new research comparing the gastrointestinal effects of milk containing only A2 type beta casein versus milk containing both A1 and A2 beta casein proteins in people who are intolerant to cows’ milk.

• Assess which dairy solution may be appropriate for clients with post-dairy digestive discomfort not caused by lactose intolerance or milk protein allergy.
Dr Andrew J Clarke is the Chief Scientific Officer of the a2 Milk Company Ltd that commercialises technology around a2 Milk® branded products, made from milk that excludes the A1 type of beta casein protein.
Milk Intolerances and allergy
*Lactase* deficient vs lactose intolerance
Lactose Intolerance and GI Inflammation
Milk Protein Intolerance
Bioactivity of fragments contained in milk protein
Beta casein variants
Beta Casomorphin-7 (BCM-7) production and activity
Beta casein variants and digestive function
Practice Implications
Milk Allergies & Intolerances

Three distinct groups of ‘adverse’ response

- Milk Protein Allergic (MPA)
  - Serious IgE mediated immune response to that may result in anaphylactic shock
  - Consumers with MPA should avoid all dairy

- Intolerant to Carbohydrate
  - Primarily lactose intolerant (LI), stems from inability to hydrolyse lactose
  - Also galactose intolerant or galactosemia, a more severe condition stemming from the inability to metabolise galactose

- Milk Intolerant or Milk Protein Intolerant
  - Present with symptoms comparable to LI and may extend to respiratory or skin irritation
  - Generally a ‘catch all’ for those not intolerant to milk carbohydrate. Reflected by consumers of goats milk who experience PD3 with cows’ milk
Lactase deficient, inflammation & lactose intolerance

- Lactose intolerance related to primary or secondary lactase deficiency is characterized by abdominal pain and distension, borborygmi, flatus, and diarrhea.
- Reported incidence of lactose or milk intolerance in consumers wide ranging:
  - Range of genetic, endoscopic and physiological tests
  - Very high incidence in some populations
  - Affected by lactose load, gut biota, and sensitivity of the gastrointestinal tract to the generation of gas and other fermentation products of lactose digestion.
- Self reported lactose intolerance (SLI) does not correspond to clinically diagnosed lactose intolerance:
  - In a lactase deficient population, approximately half of patients attending clinic with functional gastrointestinal symptoms reported intolerance to dairy products; however, SLI did not predict findings on 20 g lactose HBT (Zheng et al, 2015).
- Lactose intolerance may be a secondary effect of gastrointestinal inflammation:
  - "the majority of cases (80%) {lymphocytic colitis in pediatric population} were associated with lactase deficiency and, for the most part, gastrointestinal symptoms improved simply by treatment with Lactaid or avoidance of dairy products." (Sun et al, 2015)
  - “The risk of LI is related to the dose of lactose ingested and intestinal gas production and is increased in patients with D-IBS” (Yang et al, 2013)
Milk and GI Inflammation

- Milk has been linked to both anti-inflammatory and pro-inflammatory effects
  - “indicative of an anti-inflammatory activity in subjects with metabolic disorders and of a pro-inflammatory activity in subjects allergic to bovine milk” (Bodani et al, 2015)
- In ‘healthy’ people milk presents as anti-inflammatory (systemic, not limited to GI)
  - Emerging evidence from systematic reviews and randomized controlled trials suggests that milk product consumption does not have an adverse impact on inflammation. In fact, milk and milk products may reduce inflammation in the body by improving levels of inflammatory biomarkers. http://www.dairynutrition.ca/
- In people predisposed or with pre-existing GI conditions milk may present as pro-inflammatory
  - “The specific types of foods and beverages that can induce IBS symptoms include milk and milk containing containing products” McDermott, 2007.
- More research is required to determine what components affect who in terms of GI inflammation.
  - “Remarkably, the literature is characterized by a large gap in knowledge on bioavailability of bioactive nutrients. Future research should thus better combine food and nutritional sciences to adequately follow the fate of these nutrients along the gastrointestinal and metabolic axes”. (Bodani et al, 2015)
  - “lactose intolerance can be part of a wider intolerance to variably absorbed FODMAPs” (Deng et al, 2015)
The distilling and non-qualification of this information can lead to out of context claims...
A plethora of potential bioactive peptides are encoded in the sequence of milk proteins. Many interact with opiate receptors activity; as agonists and antagonists. Opiate activity is generally associated with anti-inflammatory effect. For their potential to be realised they must be released through enzymatic digestion and be exposed to target tissue at physiologically relevant concentrations.

(Shah et al, 2000)
Bioactive Potential of Milk Proteins (2)

- Release of a range of bioactive in physiologically relevant levels have been demonstrated to be in healthy adults (Boutrou et al, 2013)
- Gut tissue, including GALT, is exposed to the highest levels of potential bioactives, which have been observed in the small intestine of adult humans following milk protein consumption.
- The A1 type of the protein beta casein and its unique digestion product BCM-7 has been implicated in the causation of GI inflammation

Boutrou et al, 2014
Beta Casein Protein Variants

- There is approximately 2.5g of this protein in an 8oz serve
- It may be present as one of a range of natural genetic variants
- Originally all cows produced only the A2 type of beta casein
- Owing to natural genetic mutation, the A1 variant of the A2 protein appeared
- Both types have since given rise to a number of minor related “sub-variants”, such as those termed A3, B and C

Evolution of the Beta Caseins in Cow’s Milk from the Original A2 Type

Figure from: MILK PROTEIN POLYMORPHISM: DETECTION AND DIFFUSION OF THE GENETIC VARIANTS IN BOS GENUS, Annali della Facoltà di Medicina Veterinaria, Vol. XIX, 1999. Università degli Studi di Parma]
BCM-7 is released from incomplete digestion of A1

A2 types of beta casein do not release BCM-7

A1 types of beta casein and related sub-variants release BCM-7 preferentially

Beta Casein in Other Species Milk

- Human\(^1,4\), goat\(^2\) and sheep\(^3\) beta casein proteins are comparable to A2 not A1
- Only 1-10% of the human equivalent comes from in vitro digestion of human beta casein
- Internal differences mean bovine BCM-7 is 10x more potent than the theoretical human equivalent\(^5,6\)

\[
\begin{align*}
\text{Tyr}^{60}\cdot\text{Pro}^{61}\cdot\text{Phe}^{62}\cdot\text{Pro}^{63}\cdot\text{Gly}^{64}\cdot\text{Pro}^{65}\cdot\text{Ile}^{66}\cdot\text{His}^{67} & \quad \text{Bovine } \beta\text{-casein A1} \\
\text{Tyr}^{60}\cdot\text{Pro}^{61}\cdot\text{Phe}^{62}\cdot\text{Pro}^{63}\cdot\text{Gly}^{64}\cdot\text{Pro}^{65}\cdot\text{Ile}^{66}\cdot\text{Pro}^{67} & \quad \text{Bovine } \beta\text{-casein A2} \\
\text{Tyr}^{51}\cdot\text{Pro}^{52}\cdot\text{Phe}^{53}\cdot\text{Val}^{54}\cdot\text{Glu}^{55}\cdot\text{Pro}^{56}\cdot\text{Ile}^{57}\cdot\text{Pro}^{58} & \quad \text{Human } \beta\text{-casein}
\end{align*}
\]

1. embl accession X55739.1
2. embl accession AJ011019.3
3. embl accession X16482.1
A1 beta casein, BCM-7 and GI function

- BCM-7 is produced at physiological levels in healthy adult human (Boutrou et al, 2013)

- A1 consumption demonstrated to disrupt digestive function in rodents
  - Increase in inflammatory and immune markers (Ul Haq et al, 2013), Barnett et al (2014)
  - Preliminary Human Clinical Trial reports the potential link between A1 beta casein, GI inflammation and symptoms of intolerance
    - “a statistically significant positive association between abdominal pain and stool consistency was observed when participants consumed the A1 but not the A2 diet. Further studies of the role of A1 beta casein in milk intolerance are needed.” (Pal et al, 2014 and Pal et al, 2015)
A1 beta casein consumption triggers gut inflammation and immune response in mice

• A1 protein induced inflammation is BCM-7 mediated through established immune/cell pathways.

Monocyte chemotactic Protein 1

(A) IgE

(B) Total IgG

(C) IgG1

(D) IgG2

Eur J Nutr.
A1 protein derived BCM-7 implicated in triggering immune response


Fig. 1 – Effects of feeding BCM-7 and BCM-5 on inflammatory molecules in mice gut (A) intestinal myeloperoxidase, (B) monocyte chemotactic protein-1, (C) interleukin-4 and (D) histamine. The values are expressed as mean ± SEM (n = 6 animals). Different letters indicate significant differences (p < 0.05).
Mechanism of BCM-7 interaction with human cells

- A mechanisms of BCM-7’s biological interactions with neuronal and gut epithelial cells was published in 2014
- BCM-7 binds the Mu Opiate receptor which is coupled to the EAAT3 receptor
- Inhibits cysteine uptake by EAAT3 receptor, reduced substrate for antioxidant production

A1 vs A2 beta casein on lactase expression (rodent small intestine)

- A1 beta casein feeding led to an upregulation of inflammatory marker TNFα gene expression
- Conversely, this increase in inflammation corresponded to a significant down regulation of lactase gene expression in rodents
- (Data and full study submitted for publication)
Case Study: Milk Intolerance in China

• The incidence of Lactose intolerance in the Chinese population has historically been reported as high as 90% {Wang 1984}

• Commercially funded independent study by AC Nielson found
  • 85% (n=450) Chinese consumers report milk intolerance
  • 85% of these consumers drank A1 bCN-free milk and experience no PDDD

![Pie chart showing percentage of side effects after drinking a2 milk in milk intolerance consumers.]

![Bar chart showing % of side effects after drinking a2 milk in milk intolerance consumers.]

<table>
<thead>
<tr>
<th>Side Effect</th>
<th>Completely Eliminated</th>
<th>Partly Relieved</th>
<th>No change</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bloating</td>
<td>10</td>
<td></td>
<td>90</td>
</tr>
<tr>
<td>Hiccup</td>
<td>2</td>
<td>12</td>
<td>86</td>
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<tr>
<td>Diarrhoea</td>
<td>2</td>
<td>17</td>
<td>81</td>
</tr>
<tr>
<td>Nausea</td>
<td>3</td>
<td></td>
<td>97</td>
</tr>
<tr>
<td>Wind</td>
<td>3</td>
<td>23</td>
<td>74</td>
</tr>
<tr>
<td>Borborygmus</td>
<td>11</td>
<td></td>
<td>89</td>
</tr>
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• Preliminary results from double blind clinical trial (full report submitted for publication)
  • Support hypothesis that relative to A1, A2-bCN doesn’t disrupt gastrointestinal function
  • Support link of gastrointestinal inflammation as a trigger for lactose intolerance
  • Highlights the relationship between healthy gut, GSH levels & cognitive function
Post-Dairy Digestive Discomfort may lead to nutrient-poor diets

- 85% of Americans do not meet daily dairy recommendations.
- Fluid milk consumption is at an all time low.
- There is a real dairy choice for people who experience PD3 not attributable to LI.

Thank You

- Further references can be found at [www.betacasein.net](http://www.betacasein.net) and [www.betacasein.org](http://www.betacasein.org)
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