Value Added Co-product from *Jatropha* Biodiesel Production Process






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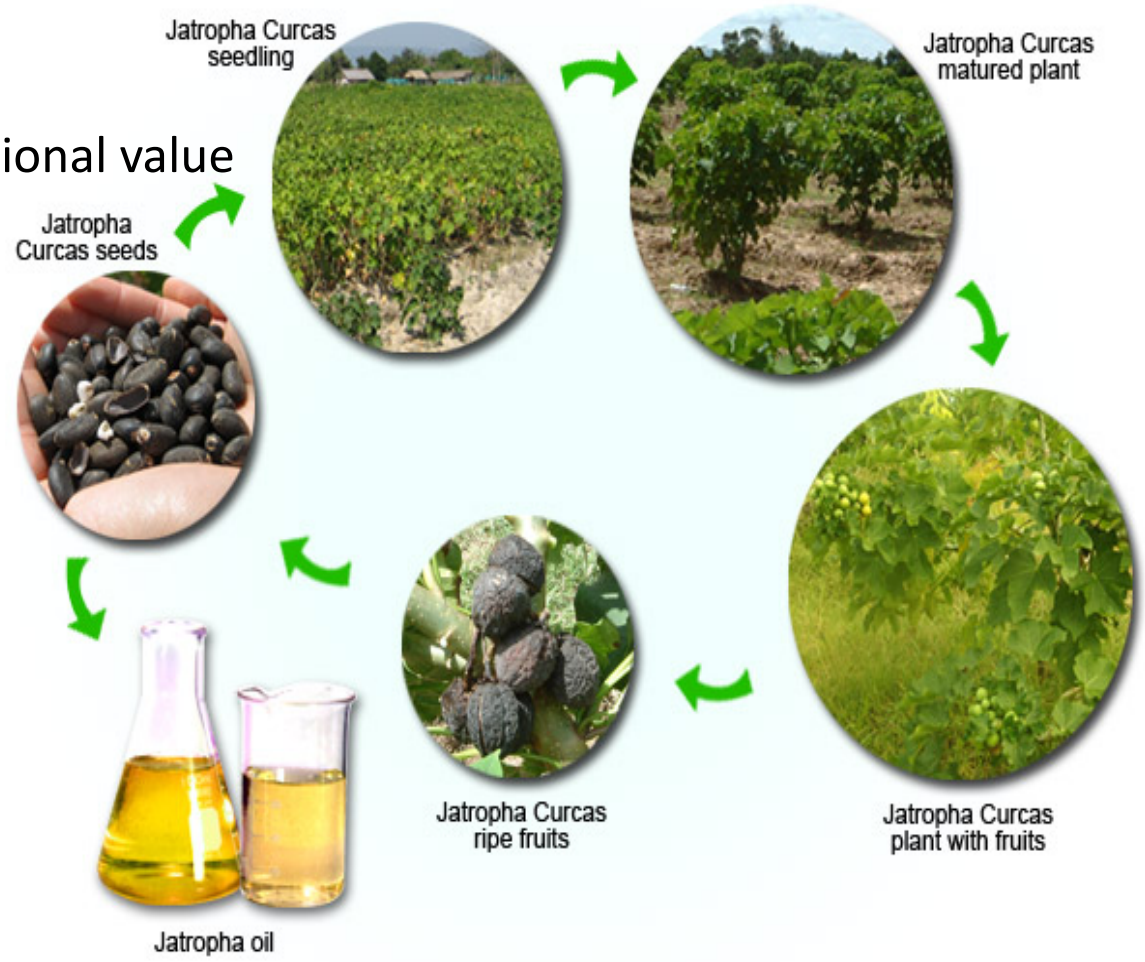
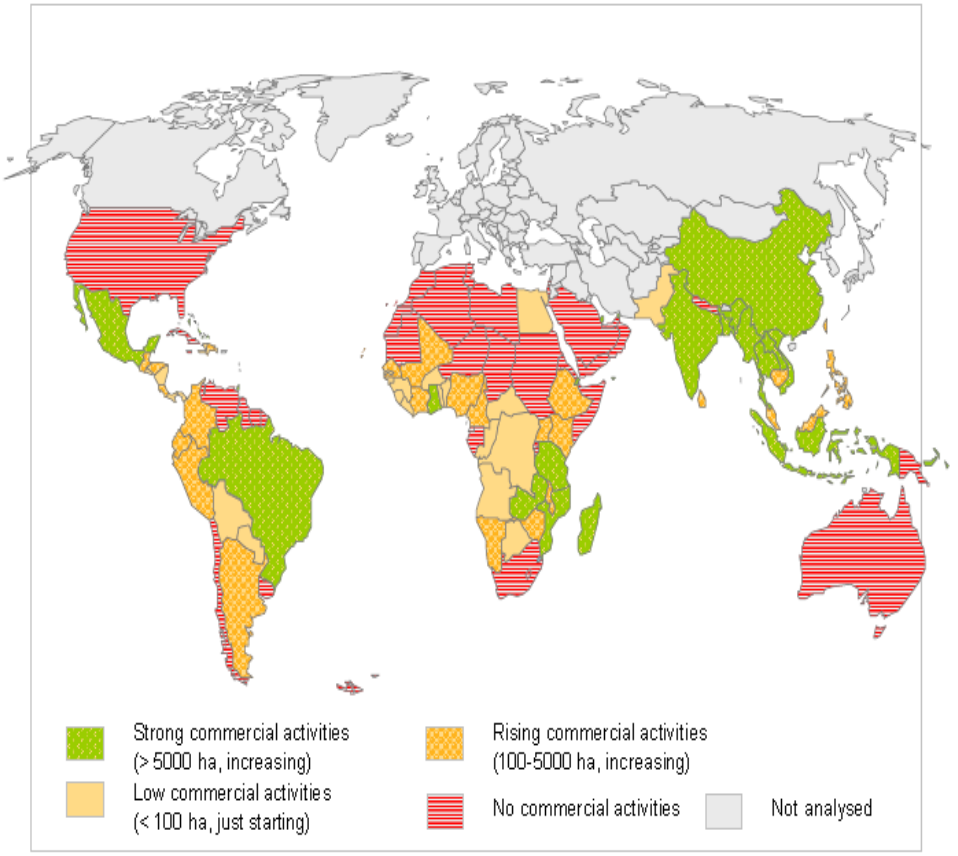


Jatropha curcas

- **Origin:** Central American countries
- **Common names:** physic nut, pig nut, purging nut etc.
- **Distribution and habitat:**
  - Dry regions of tropics
  - Rainfall : 11.8" – 39.4"
  - Altitude: 0-500 m
  - Annual temp. (Avg) : 65 F
- **Botanical features**
  - Euphorbiaceae family
  - Small tree or shrub
  - When cut exudes white late
  - Grows to 9 -16 feet
  - Yield (5-7 mt/h)



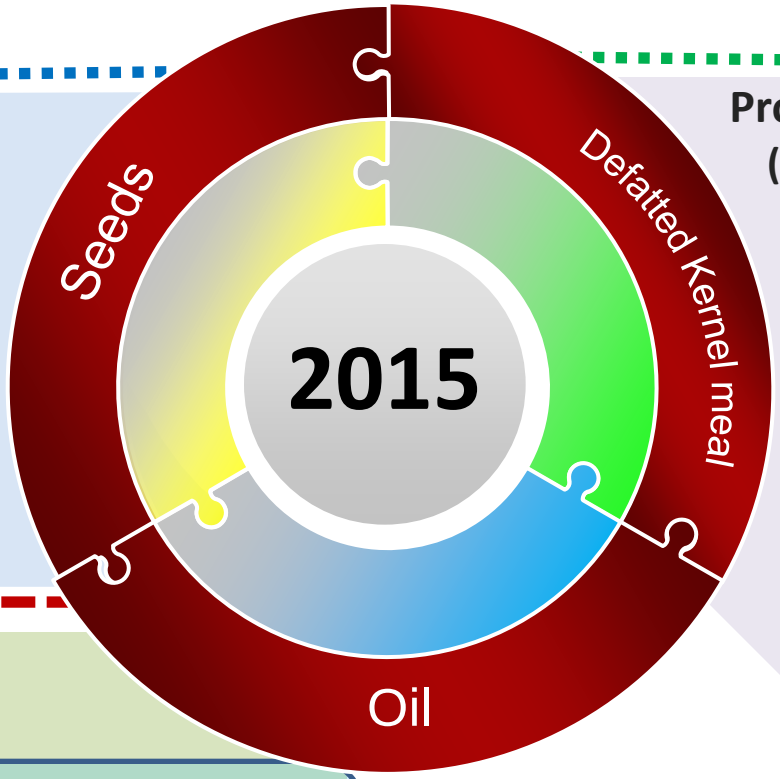
Multipurpose uses

Future *Jatropha* Production

Production of seeds

~28 billion pounds

Considering (28% oil)



Production of:

(a) Defatted kernel meal

7.4 billion pounds

Considering:

- ***63:37 = Kernel:shell***
- ***58% oil in kernel***

(b) Defatted Seed cake

20.16 billion pounds

Considering:

- ***28% oil recovery from seeds***

Production of oil

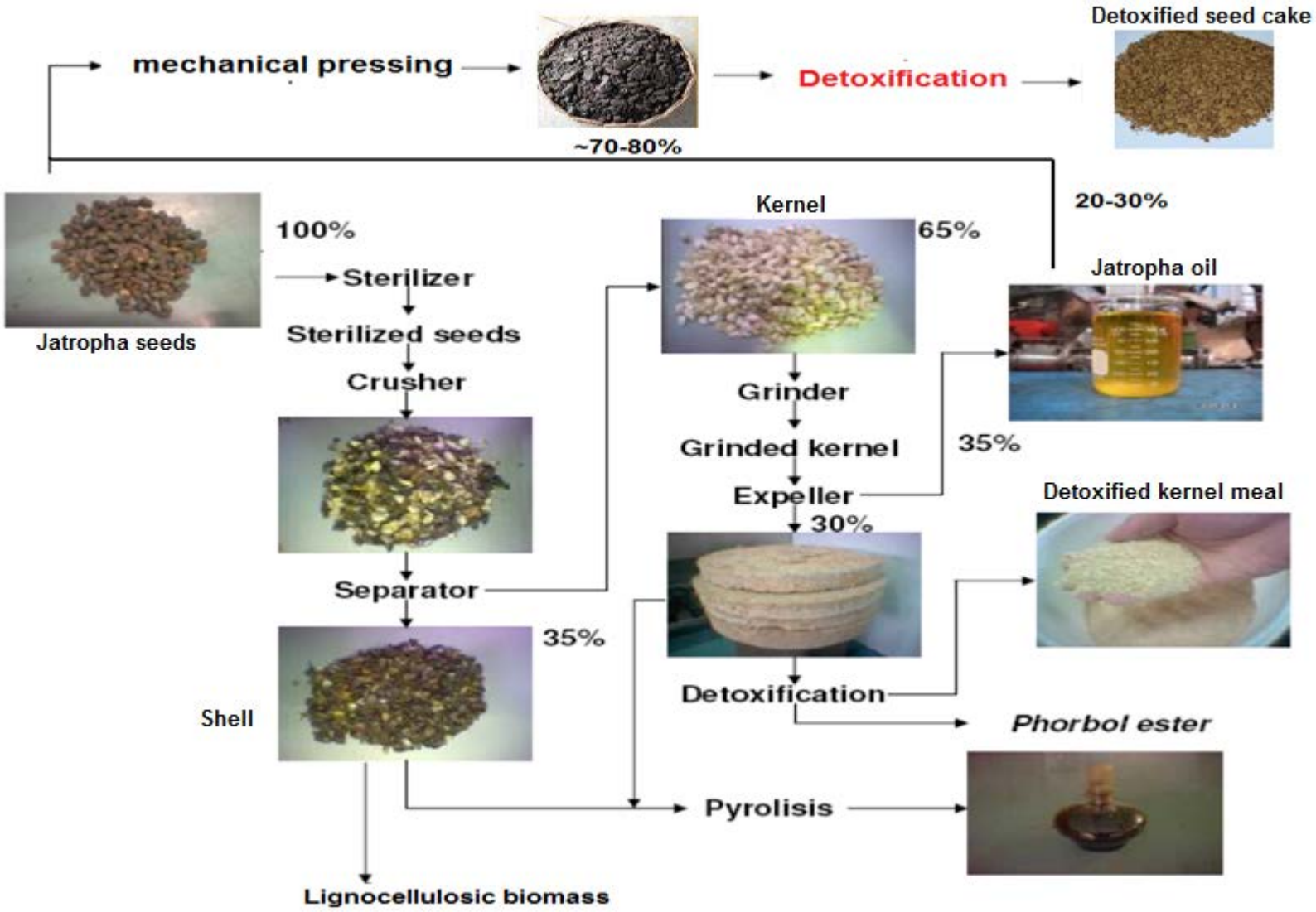
7.84 billion gallons

Considering

- ***248 gallons oil/acre***
- ***Cultivation*** **-31.63 million acres**

Source: GEXSI market study on Jatropha (2008)

Jatropha curcas Processing



Source: H.P.S. Makkar and K. Becker, University of Hohenheim, Stuttgart, Germany

How Good is *Jatropha* as Fish Feed?

Feedstock	Market share (%)	Energy (MJ/kg DM)	Protein (%)	Fibre (NDF %)	Antinutritional factors	Detoxification treatments
Soybean meal	70	12	50 -53	6	Trypsin inhibitor, phytic acid, lectins, bitter taste, oligosacharides	Heat treatment and solvent extraction
Rapeseed Meal	12	12	39	12-14	Glucosinolates	Controlled feeding studies
Sunflower meal	6	9.5	37	15	Chlorogenic acid	Washing
Cotton meal	6	11.5	40	15	Gossypol	Controlled feeding levels, breeding, solvent extraction
Jatropha kernel meal	0	18	55-64	10	Phorbol esters, curcin, trypsin inhibitor, lectin, saponin, phytate	Chemical and solvent

Source: H.P.S. Makkar and K. Becker, University of Hohenheim, Stuttgart, Germany

Chemical Composition of Toxic and Nontoxic Defatted *Jatropha curcas* Kernels

Parameters	Variety (*Jatropha Curcas*)	
	Toxic	Nontoxic
Crude protein (%)	56-65	63.8
Lipid (%)	1.5	1.0
Ash (%)	9.6	9.8
Gross energy (MJ/kg)	18.2	18
Neutral Detergent Fibre (%)	9.0	9.1

Non toxic *Jatropha platyphyllla* seeds

Toxic *Jatropha curcas* seeds

Source: H.P.S. Makkar and K. Becker, University of Hohenheim, Stuttgart, Germany

Quality of Amino Acid Profile (g/100 g protein)

Amino acids	Cape Verde genotype	Nontoxic Mexican genotype	Protein Isolate	Soybean	Essential amino acid requirement			FAO reference protein
					Fish	Chick	Pig	
Lysine	4.28	3.4	3	6.08	4.1-6.1	6.1	4.7	5.80
Leucine	6.94	7.5	7.08	7.72	2.8-5.3	6.7	4.6	6.60
Isoleucine	4.53	4.85	4.47	4.62	2.0-4.0	4.4	4.6	2.80
Methionine	1.91	1.76	1.66	1.22	2.2-6.5[a]	4.4[#]	3.0[#]	2.50
Cystine	2.24	1.58	1.34	1.70	5.0-6.5	-	-	
Phenylalanine	4.34	4.89	5.42	4.84	5.0-6.5[b]	7.2*	3.6*	6.30
Tyrosine	2.99	3.78	3.2	3.39	-	-	-	
Valine	5.19	5.3	5.18	4.59	2.3-4.0	4.4	3.1	3.50
Histidine	3.3	3.08	3.51	2.50	1.3-2.1	1.7	1.5	1.90
Threonine	3.96	3.59	3.56	3.76	2.0-4.0	3.3	3	3.40

[a] requirement varies depending on the amount of cystine in the diet

[b] requirement varies depending upon the amount of tyrosine in the diet

[#] in the absence of cystine

*in the absence of tyrosine

Source: H.P.S. Makkar and K. Becker, University of Hohenheim, Stuttgart, Germany

Antinutrient/toxic Constituents Present in Toxic and Nontoxic Defatted *Jatropha curcas* Kernel meal*

Component	Variety	
	Toxic	**Nontoxic**
Phorbol ester (PEs; mg/g)	3.00	ND to 0.11
Total phenols (% tannic acid equivalent)	0.36	0.22
Tannins (% tannic acid equivalent)	0.04	0.02
Phytates (% dry matter)	9.40	8.90
Saponins (% diosgenin equivalent)	2.60	3.40
Trypsin inhibitor (mg trypsin inhibited per g sample)	21.30	26.50
Lectins (1/mg of meal that produced haemagglutination per ml of assay medium)	102	51

*Expressed on dry matter basis

Source: H.P.S. Makkar and K. Becker, University of Hohenheim, Stuttgart, Germany

Toxicity of *Jatropha* Seeds

Mice: LD50 27 mg/kg Bd. wt.

Goat and sheep: 0.05 g/kg/day in diet

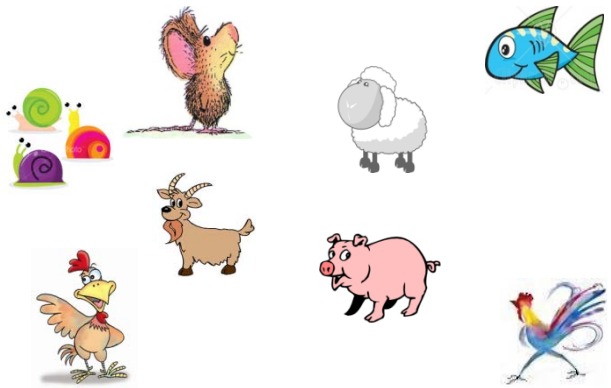
Chicks: 0.1% seed in diet

Molluscs: 3 ppm of ethanol extract

Fish: 15 ppm PEs in diet

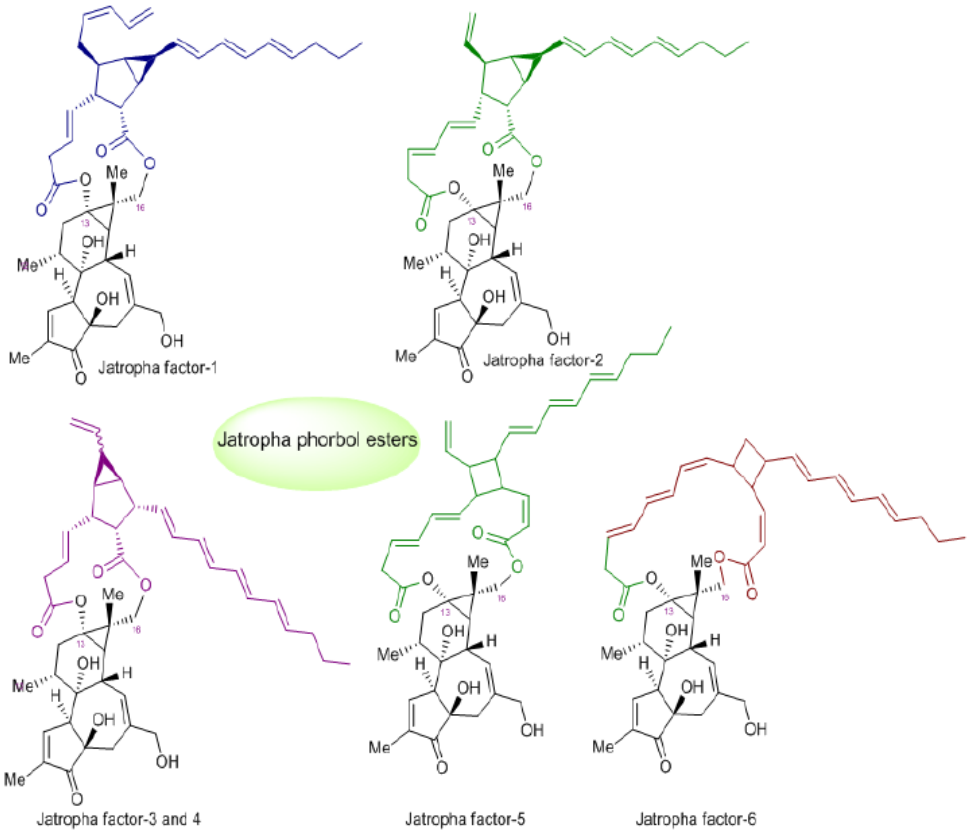
Pig: 0.8 mg/g PEs in diet

Humans: No authenticated studies



Source: Haas et al., 2002; Source: H.P.S. Makkar and K. Becker, University of Hohenheim, Stuttgart, Germany

- **6 types of phorbol esters.**
- **Heat stable, Tumour promoters**
- **After pressing: 70% of PEs in oil + 30% of PEs in seed cake**
- **Quantification: HPLC method, LCMS**



Detoxification of *Jatropha* Cake/Kernel Meal

- **Criteria:**

1. **Favourable:** Process should be fast, cost effective, phorbol esters (PEs) should be reduced to undetectable level in HPLC, exhibit no toxicity in bioassays and in animal trials.
2. **Not favourable:** time consuming, PEs reduced below or level similar to nontoxic genotypes of Jatropha containing PEs.

- **Current detoxification methods to remove PEs**

1. **Solvent** – effective, time consuming
2. **Chemical** – effective, bit harsh on the amino acid profile
3. **Enzymatic** – promising but not completely effective
4. **Microbial** - time consuming, potentially effective

Effect of Detoxification* on *Jatropha* Cake/Kernel Meal

Parameter		Untreated Jatropha kernel meal	Treated Jatropha Kernel meal	Soy bean meal	FAO**
Protein (% DM basis)		64.0	63.0	50-53	
Amino acid (g/16 g N)	Methionine	1.84	1.55	1.22	2.50
	Cystine	1.51	1.36	1.70	
	Lysine	3.60	3.36	6.10	5.80
Protein digestibility % (pepsin + pancreatin)		95.2	85.8	91-95	

* WIPO Patent Application WO/2010/092143

**Ref. Protein for growing child

Source: H.P.S. Makkar and K. Becker, University of Hohenheim, Stuttgart, Germany

Is Detoxified* *Jatropha* Kernel Meal Fit for Aqua Feed


Common carp

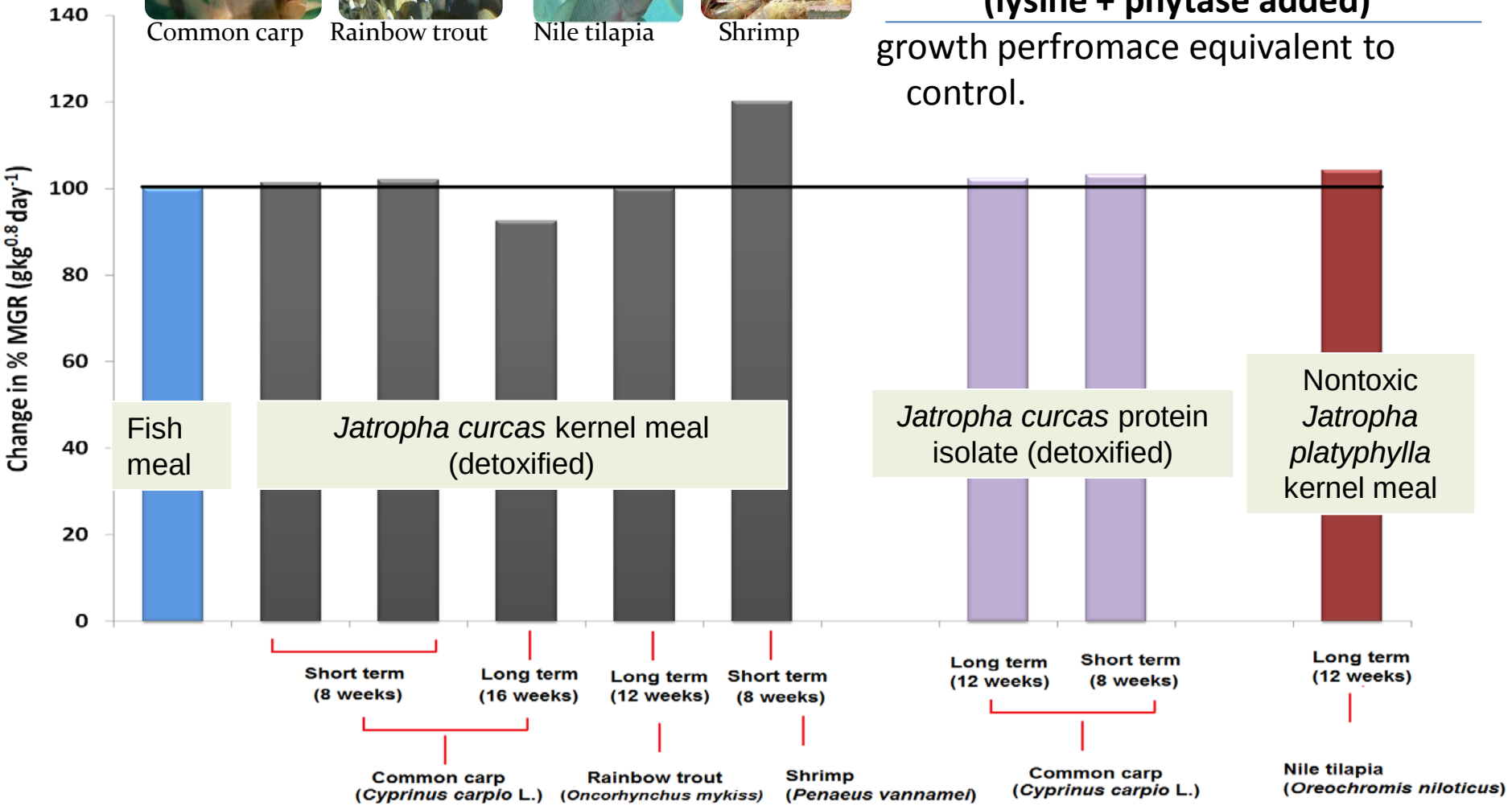

Rainbow trout


Nile tilapia


Shrimp

Upto 50% protein replacement in fishmeal based diet (lysine + phytase added)

growth perfromace equivalent to control.

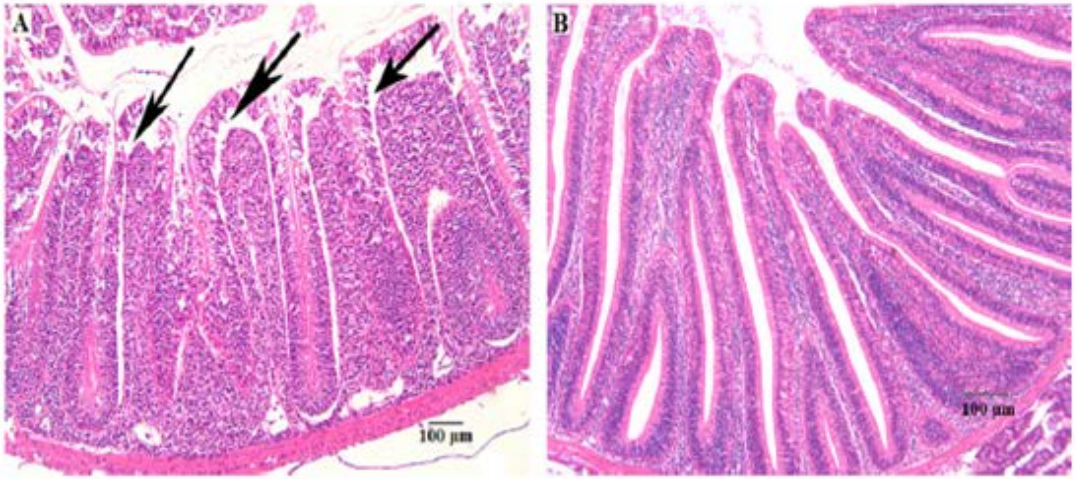


MGR: Metabolic growth rate

* WIPO Patent Application WO/2010/092143

Source: H.P.S. Makkar and K. Becker, University of Hohenheim, Stuttgart, Germany

Histopathology in Fish (*Cyprinus carpio*)



Stomach villi
(Jatropha kernel meal)

Stomach villi
(Detoxified Jatropha kernel meal)

NO significant changes observed when fed on detoxified * Jatropha kernel meal when compared to Fish meal based diet.

- ✓Palatability
- ✓Nutrient digestibility and digestive enzymes
- ✓Growth
- ✓Energy metabolism

- ✓Growth hormone and IGF encoding genes
- ✓Clinical markers enzymes

- ✓Gut morphology

- ✓No toxicity in - liver, kidney, intestine, stomach, heart

- ✓No toxicity -serum biochemical characteristics : glucose and cholesterol levels, haemoglobin, haematocrit and triglyceride

* WIPO Patent Application WO/2010/092143

Source: H.P.S. Makkar and K. Becker, University of Hohenheim, Stuttgart, Germany
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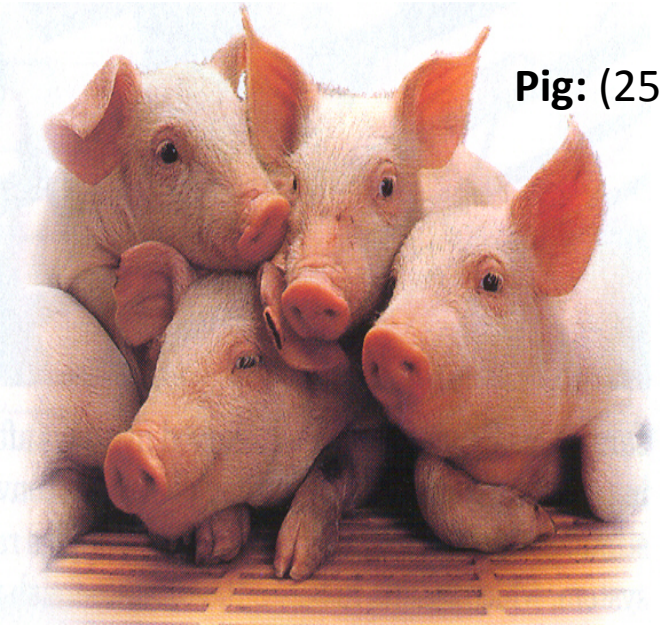
Detoxified* *Jatropha* Meal in Animal Feed

Poultry: (10% and 20% in maize + soybean meal based diet)

- Digestibility of AA from DJKM in young turkeys is very high in comparison with data from other feeds



Pig: (25 and 50% replacement of soybean iso-nitrogenous diet)

- No histopathological or serum biochemical parameters were altered
- Detoxified Jatropha meal could replace up to 50% of soybean meal protein with no significant change in growth and feed conversion ratio.

* WIPO Patent Application WO/2010/092143

Source: H.P.S. Makkar and K. Becker, University of Hohenheim, Stuttgart, Germany

Summary

- **Jatropha kernel meal/seed** cake is rich in **protein**
- Kernel meal/seed cake **contain phorbol esters as toxic constituent**
- Kernel meal/seed cake **can be detoxified**
- **Detoxified meal** has a **high acceptability** when fed to animals (**fish**, poultry and pig)
- Inclusion of detoxified meal did not affect the growth and health of fish.

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Information Sources

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2. Vikas Kumar, PhD thesis submitted to the University of Hohenheim (2010).
3. Makkar HPS and Becker K. Method for detoxifying plant constituents (WIPO Patent Application WO/2010/092143)
4. Nitayavardhana, S., and Khanal, S. 2011. Biodiesel-derived crude glycerol bioconversion to animal feed: A sustainable option for a biodiesel refinery. Bioresource Technology, 102, 5808-5814.