

**Invited Review**

## **Resistin: molecular history and prognosis**

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**Abstract** Obesity and diabetes have reached epidemic proportions worldwide. The antidiabetic thiazolidinedione (TZD) drugs are insulin-sensitizing agents now widely used in the treatment of type 2 diabetes. TZDs are ligands for the nuclear hormone receptor peroxisome proliferator activated receptor  $\gamma$ , which is a master regulator of adipogenesis and adipocyte metabolism. The molecular mechanisms by which TZDs improve insulin sensitivity have not been fully identified. Here we consider a novel secreted factor first identified as a TZD-suppressible gene in mouse adipocytes, called resistin, and discuss what is currently known about resistin regulation and function in mouse and human.

**Keywords** Resistin · Obesity · Type 2 diabetes · Insulin resistance · Thiazolidinediones

#### **Abbreviations**

*FIZZ* Found in inflammatory zone

*LPS* Lipopolysaccharide

*NEFA* Nonesterified fatty acid

*PPAR* Peroxisome proliferator activated receptor

*RELM* Resistinlike molecule

*SNP* Single-nucleotide polymorphism

*TNF* Tumor necrosis factor

*TZD* Thiazolidinediones

*WAT* White adipose tissue

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## **Insulin resistance and thiazolidinediones**

Insulin resistance, the inability of target tissues to respond normally to insulin, is central to the pathophysiology of type 2 diabetes [1], but its molecular basis remains poorly understood. Classically, the explanation for the cause of insulin resistance has relied on the effects of elevations in serum nonesterified fatty acids (NEFAs) on metabolism [2]. Increases in NEFAs favor increases in fatty acid oxidation, which elevates acetyl CoA production in mitochondria.

This in turn inhibits pyruvate dehydrogenase, the rate-limiting enzyme for glucose oxidation, and interferes with glucose utilization, a mechanism known as the Randle effect [3]. In addition, NEFAs suppress pancreatic  $\beta$ -cell insulin secretion and stimulate hepatic glucose output [3].

More recently it has been recognized that, in addition to shifting substrate utilization away from glucose, NEFAs contribute to insulin resistance in muscle by decreasing insulin stimulated glucose uptake [4]. This is mediated in part by increases in intracellular diacylglycerol concentrations, leading to activation of protein kinase C- $\theta$ , which increases insulin receptor substrate 1 Ser-307 phosphorylation [5]. Activation of insulin receptor substrate 1 associated phosphatidylinositol 3-kinase activity is decreased, and insulin-stimulated glucose transport is reduced. Thus insulin resistance in obesity is partly a consequence of elevated NEFA levels [3, 6].

Further elucidation of the molecular basis of insulin resistance has come through studying a new class of synthetic drugs called the thiazolidinediones (TZDs), including rosiglitazone and pioglitazone. TZDs were identified clinically as drugs that lower blood glucose and insulin levels, leading to improved insulin sensitivity in type 2 diabetics [7, 8]. TZD treatment lowers NEFAs, in addition to glucose, but patients also typically gain weight. This is a paradox, since weight gain per se should exacerbate diabetes by worsening obesity and insulin resistance [9, 10]. In 1995 the observation that TZDs were high-affinity ligands for the nuclear receptor peroxisome proliferator activated receptor (PPAR)  $\gamma$  [11] suggested that the mechanism of TZD improvement of insulin resistance involve PPAR $\gamma$ .

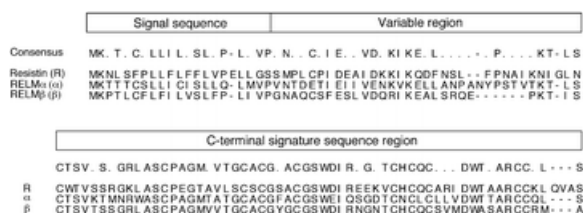
However, this raised another paradox [8, 12]. PPAR $\gamma$  is a master regulator of adipogenesis [13, 14] and expressed at highest levels in adipose cells [8]. However, TZD-associated improvement in insulin sensitivity was associated with improved glucose disposal into muscle [15]. Furthermore, in mice lacking fat TZD treatment failed to lower glucose levels, indicating that their glucose lowering effects required the presence of adipose tissue [16]. Intriguingly, TZD treatment lowered NEFA levels in this model, thus suggesting that decreases in serum NEFAs levels are not sufficient to improve insulin resistance.

The discovery over the past few years that adipose tissue secretes a number of hormones and bioactive factors suggests a way to explain these observations and resolve the apparent paradoxes associated with adipose PPAR $\gamma$  as the molecular target of TZDs [8]. The perception of adipose tissue as merely a passive energy storage organ has changed to reflect a new appreciation for its roles as an endocrine organ critical in regulating whole body metabolism

[17, 18, 19]. A number of adipocyte-derived hormones, notably leptin, but also including tumor necrosis factor (TNF)  $\alpha$ , adiponectin/ACRP30, interleukin 6, plasminogen activating inhibitor 1, adipsin, and steroid hormones, among others, have been identified [18]. Leptin, TNF $\alpha$ , adiponectin, and interleukin 6 are all associated with and contribute to insulin resistance [20]. In fact, leptin, TNF $\alpha$  [8], and adiponectin [21] are regulated by TZD treatment in a manner consistent with improvements in insulin resistance.

## Discovery of resistin

Although TZDs have been shown to regulate a number of factors contributing to insulin resistance and regulation of metabolism, it is plausible that hitherto unidentified TZD-regulated genes also contribute to their antidiabetic and insulin-sensitizing effects. To test this possibility a subtractive screen was performed on 3T3-L1 adipocytes, a widely used cell culture model of white adipocytes [14, 22]. This screen led to the identification of a novel messenger RNA that is downregulated by rosiglitazone [23], a finding later recapitulated by a genomic screening approach using Affymetrix gene chips [24]. The novel gene encoded for a 114 amino acid polypeptide with an amino terminal signal sequence, suggesting that it is a secreted molecule (Fig. 1). Named resistin, the protein was found to contain a pattern of 11 cysteine residues in a unique motif (X11-c-x8-c-x-c-x3-c-x10-c-x-c-c-c-x9-cc-x6) [23]. Similar cysteine rich motifs are common among secreted growth factors [25]. In vitro studies showed that resistin is induced during adipocyte differentiation and, as suggested by its signal sequence, is secreted into media [23]. In mice resistin is highly and specifically expressed in white adipose tissue (WAT) [23]. Mouse studies also showed that resistin is detectable in serum, demonstrating that it can circulate in blood. Treatment of mice with TZDs lowered resistin protein levels, similar to the effects observed in vitro [23].



**Fig. 1.** The murine family of resistin and resistin-like molecules (*RELMs*). Sequences for resistin, *RELM $\alpha$*  and *RELM $\beta$*  were aligned using the clustal method of DNASTAR. The consensus sequence is shown *above the alignment*. Location of the signal sequence, variable region, and C-terminal signature sequence region are indicated

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In both environmental (high fat diet induced) and genetic (leptin-deficient, *ob/ob*; leptin receptor-deficient, *db/db*) models of obesity in mice resistin serum levels were elevated [23]. Additionally, both WAT mRNA and serum protein levels of resistin dropped during fasting and increased during refeeding. Regulation of resistin by nutritional status is interesting because it parallels that of leptin [26]. Although correlative, these observations pointed towards a potential role for resistin in obesity.

To address more directly whether resistin contributes to obesity-associated insulin resistance its protein levels were manipulated in vitro and in vivo. In mice with diet-induced obesity, immunoneutralization of resistin resulted in a 20% drop in blood glucose, and improved insulin sensitivity as measured by insulin tolerance testing [23]. Conversely, treatment of wild-type mice with purified recombinant FLAG-tagged resistin protein led to modest glucose intolerance. Treatment of 3T3-L1 adipocytes with resistin protein impaired insulin-stimulated glucose uptake [23].

Several groups subsequently published findings independently describing the identification of resistin by various methods. Rajala and colleagues [27] isolated resistin as an adipocyte-specific transcript in a subtractive screen technique comparing adipocytes during different stages of differentiation. Kim and colleagues [28] identified adipose tissue-specific secretory factor (resistin) in rats using a cDNA microarray screen for novel adipocyte specific genes induced during differentiation. These groups confirmed that resistin is regulated by fasting and refeeding [27, 28] with the most pronounced effects seen in perirenal fat [27]. Resistin also inhibited the differentiation of 3T3-L1 preadipocytes and was strongly induced in streptozotocin-diabetic mice following treatment with insulin [28]. In Sprague-Dawley rats a gene expression profile screening approach identified resistin as one of the most strongly upregulated genes following a week of 60% high fat diet [29]. Recently resistin was identified with a proteomic approach using liquid chromatography based separation, followed by tandem mass spectroscopy as one of a number of proteins secreted by mature adipocytes, as compared to those identified from preadipocytes [30].

Together these initial studies identified resistin in screens using a variety of techniques on both in vitro and in vivo (rodent) model systems of adipogenesis and obesity. They also show agreement that resistin is an adipose-specific secreted molecule that is induced during adipocyte differentiation, and is strongly regulated by nutritional status in rats and mice.

Database searching using the resistin cDNA sequence revealed the existence of a family of related proteins called resistinlike molecules (RELMs; Fig. 1) [31]. These proteins were independently identified as the found in inflammatory zone (FIZZ) family following the observation that FIZZ1/RELM $\alpha$  is upregulated in a mouse model of allergic pulmonary inflammation [32]. As with resistin (FIZZ3), the two mouse homologues, named RELM $\alpha$  (FIZZ1) and RELM $\beta$  (FIZZ2), are highly cysteine rich and contain signal sequences [31]. However, each has a unique tissue-specific distribution: RELM $\alpha$  is prevalent in the stromal-vascular fraction of adipose tissue [27] and lung [32] while RELM $\beta$  is found in the proliferative epithelial cells of the colon [31]. All three mouse homologues have been found to be secreted when expressed in 293T cells in vitro [31].

## Biochemistry of resistin

Resistin has been found to be secreted as a dimer [25, 27]. Surprisingly, while RELM $\beta$  shares the same cysteine residue pattern and is also secreted as a dimer, RELM $\alpha$  lacks the most amino terminal cysteine (Cys-26 in resistin) and is secreted as a monomer [25]. When the Cys-26 residue of resistin is mutated to an alanine, it is secreted as a monomer, suggesting that this residue is critical for dimerization [25]. These results suggest that the ten conserved cysteines form a common monomeric structural motif, and that dimerization is determined by the presence or absence of the most amino terminal cysteine. In support of this hypothesis are the findings of pulse-chase experiments in which the monomer form of resistin appears to partially fold prior to dimer formation [27]. Pulse-chase experiments also reveal that resistin has a relatively slow kinetics of secretion, with a half-life of over 5 h [27]. The functional relevance of dimerization has yet to be determined, but it is hypothesized that it is critical to biological function, based on previous examples of other growth factors [25]. Recent studies suggest that resistin forms higher order multimeric complexes with other resistin dimers or with the other RELMs [33]. This is particularly interesting given the precedent of adiponectin, which forms higher order structures that appear to regulate its function in insulin sensitivity [34, 35, 36].

## Regulation of resistin gene expression

The discovery of resistin as a new adipose-specific secreted molecule implicated in diabetes and obesity generated excitement in the field. A number of groups have investigated regulation

of resistin in animal models of diabetes and obesity and by a variety of agents known to modulate insulin sensitivity. These results are discussed below and summarized in Table 1. [Table 1. will appear here. See end of document.]

## Regulation by TZDs, obesity, and nutritional status

Several early studies sought to confirm the regulation of resistin by TZDs in 3T3-L1 adipocytes. Consistent with the findings of Stepan et al. [23], resistin was found to be downregulated not only by rosiglitazone but also by darglitazone [37] and troglitazone [38]. This suggests that downregulation of resistin is broadly a characteristic of TZDs and is not ligand restricted. Even when adipocyte differentiation is driven by a constitutively active PPAR $\gamma$ , rosiglitazone is still capable of downregulating resistin expression, indicating that the negative regulation either can functionally overcome activated PPAR $\gamma$  or is not directly PPAR $\gamma$  dependent [24]. The observation that treatment of 3T3-L1 adipocytes with 10  $\mu$ M rosiglitazone for 12 h failed to decrease resistin protein in media is not surprising [27] given the approximately 24-h half-life of resistin [39].

There is less consensus regarding the effect of TZD treatments on resistin expression in vivo. Moore et al. [40] investigated the effect of chronic rosiglitazone treatment on *db/db* female mice and found that after 11 weeks of treatment resistin mRNA in parametrial WAT is decreased 72%, consistent with the initial reports. Lu et al. [41] found that rosiglitazone treatment for 4 weeks lowers resistin by 60% in rats. By contrast, Way et al. [42] found that treatment of *ob/ob* male mice with rosiglitazone or PPAR $\gamma$  agonists MCC-555 or GW1929 actually *increases* resistin expression several-fold in WAT following a treatment lasting 7–10 day. These treatments were confirmed to have their expected effects on other known target genes of PPAR $\gamma$  such as FATP and PEPCK and to have their expected effects in lowering serum glucose and increasing insulin sensitivity [42]. Fukui and Motojima [43] similarly found that an 8-day treatment with pioglitazone or troglitazone slightly induces resistin in both lean C57BL and KK and obese KKA<sup>y</sup> and *db/db* mice. This suggests that downregulation of resistin is not required for the antidiabetic effect of TZDs in all model systems, and that the length of treatment is critical in determining the regulation of resistin by TZDs.

Supporting the association between resistin and obesity-induced insulin resistance is the observation that resistin expression in visceral fat is 15-fold higher than in subcutaneous fat [44]. Visceral fat has been identified as a risk factor for insulin resistance, and increased

levels of visceral fat are correlated with decreased sensitivity to insulin [1, 44, 45]. In studies of c-Jun amino-terminal kinase knockout mice serum resistin protein levels have been found to be significantly decreased, consistent with their improved insulin sensitivity [46]. However, both Way et al. [42] and Fukui and Motojima [43] observed that resistin expression is decreased in other genetic models of obesity in mice, including *ob/ob*, *db/db*, *tub/tub*, and *KKA<sup>y</sup>* mice, as compared to their lean counterparts. Similar changes have been observed in *ob/ob* mice in other studies [20, 27]. Additionally, in several strains of FVB mice diet-induced obesity or gold thioglucose-induced obesity either had no effect or regulated resistin [47]. In Fischer 344 rats resistin expression increased in proportion to body weight increase, but no change in insulin resistance was observed [48]. Furthermore, in rat models of insulin resistance induced by fructose feeding, a lean model of insulin resistance, resistin expression was decreased [49].

One possible explanation for reduced adipocyte resistin gene expression in models of obesity is that individual fat cells downregulate resistin mRNA levels, but the increase in the total adipose tissue mass nevertheless leads to an increase in the circulating levels of resistin protein. Another theoretical possibility is that in obesity either the stability or clearance of resistin is altered, leading to elevated levels despite decreased expression. Obesity-related modifications in the resistin protein might have different activities, as has been shown to be the case for another adipocyte-derived hormone, adiponectin [35].

## **Regulation by insulin and glucose**

Treatment of 3T3-L1 adipocytes with insulin for 48 h was found to significantly reduce resistin mRNA levels, with a 50% reduction occurring within physiological levels [37]. Similar magnitude of effects were seen in both resistin message and protein levels in another study [38]. These results in vitro are in contrast to the results previously described in vivo, in which insulin administration induced resistin [28]. Conversely, high glucose levels, both in studies of 3T3-L1s in vitro [38] and in vivo during hyperglycemic/euinsulinemic clamp studies of rats [27], showed strong induction of resistin expression. One explanation for these apparently contradictory results is that acute elevations in glucose or insulin in vitro are not sufficient to mimic the effects of hyperglycemia, hyperinsulinemia, or insulin resistant states on resistin expression in vivo.

## Glucocorticoids

Several groups investigated whether resistin expression was affected by glucocorticoids, long associated with insulin resistance [50]. Resistin is upregulated several-fold following dexamethasone treatment in vitro [37, 38], but whether this effect is independent of dexamethasone-stimulated adipogenesis is unclear. Additionally, treatment of mice with dexamethasone, which leads to hyperinsulinemia but not fasting hyperglycemia, induces resistin in WAT 70–80% in the absence of elevations in TNF $\alpha$  [38]. While only correlative, this suggests that resistin is associated with and contributes to glucocorticoid induced insulin resistance [38]. Removal of the adrenals has been shown to improve insulin sensitivity. Makimura et al. [51] investigated the effect of adrenalectomy on mRNA levels of resistin and adiponectin in *ob/ob* mice. While adiponectin levels increased in treated animals, resistin levels did not decrease and surprisingly were positively correlated with those of adiponectin [51].

## Proinflammatory stimuli

Proinflammatory cytokines have been implicated in insulin resistance associated with adipose tissue, particularly TNF $\alpha$ , and interleukin 6 [20]. Surprisingly, TNF $\alpha$  treatment yields moderate [27] to strong [38, 52] downregulation of resistin in 3T3-L1 adipocytes, and interleukin 6 treatment has no effect on resistin levels in vitro [27]. Bacterial lipopolysaccharide (LPS) is a potent proinflammatory stimulus which can cause hyperglycemia and insulin resistance. LPS treatment of rats induces resistin expression 66% in white adipose tissue and white blood cells as well as in 3T3-L1 adipocytes, according to Lu et al. [41]. However, Rajala et al. [27] found that LPS treatment of 3T3-L1s has no effect on resistin expression, and LPS treatment of FVB mice actually downregulates resistin in adipose tissue. Therefore the regulation of resistin by this factor remains unclear.

## Other regulators of resistin expression

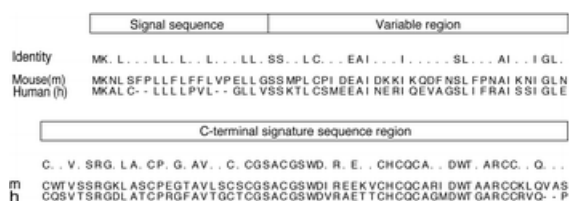
Several other metabolic states or factors associated with insulin resistance have also been investigated. Resistin is elevated by hyperprolactinemia [53] and testosterone [53] but not angiotensin 2 or growth hormone [52]. However, resistin is strongly induced by growth hormone administration in growth hormone deficient spontaneous dwarf rats, known to have very low TNF $\alpha$  levels [54].  $\beta_3$ -Adrenoreceptors have also been implicated in insulin resistance,

and the  $\beta_3$ -agonist isoproterenol reduced resistin levels in vitro by 20%, reversible by the  $\beta$ -antagonist propranolol [55]. Furthermore, this effect seems to be mediated via a protein kinase A dependent pathway since it is reproduced with cholera toxin or forskolin treatment [55]. However, other groups found no effects with the  $\beta_3$ -agonists BRL37344 [37] or BRL-35135 [40], and thus the regulation by this class of drugs remains unclear. Additionally, resistin is suppressed by epinephrine and somatotrophin [38].

In summary, there is consensus that resistin is regulated by adipogenesis and by fasting and refeeding, and is downregulated at the RNA level by obesity. However, the association between resistin levels and insulin resistance in various models is controversial since resistin is not elevated in all models of insulin resistance. While the reported regulation of resistin by other factors is intriguing, no underlying principle is apparent, and the physiological relevance of most of these effects remains to be determined.

## Human resistin

Based upon the studies of resistin regulation and function in rodents a great deal of interest was generated in determining how the rodent studies apply to the mechanism of type 2 diabetes in humans. Translating the findings of murine resistin biology to human pathophysiology has been complicated, however, by the fact that only two human homologues have been found for the three mouse genes [31]. Overall, human resistin is only 53% identical with its murine counterpart, but identity is highest in the C-terminal signature sequence region [31] (Fig. 2). Studies of human resistin thus far fall broadly into two categories: genetic studies analyzing association of the human resistin locus with diabetes or obesity, and expression studies measuring resistin mRNA or protein levels in these states. As with the murine studies, the literature is divided with reports both for and against a role for resistin in human obesity or diabetes.



**Fig. 2.** Comparison of mouse and human resistin. Sequence identity is shown above the alignment. Location of the signal sequence, variable region, and C-terminal signature sequence region are indicated

## Expression studies

Surprisingly, unlike murine resistin, human resistin appears to be expressed at low levels in adipocytes, but is readily detectable in mononuclear blood cells [56, 57]. Monocytes isolated from peripheral blood and differentiated in vitro to macrophages show a fourfold induction of resistin mRNA; treatment of these cells with rosiglitazone downregulates resistin expression 80% [58]. These results suggest that despite differences in tissue expression pattern the mechanism of TZD regulation of human and mouse resistin is conserved. Nagaev and Smith [56] detected only a very low level of resistin in fat or muscle samples from 42 individuals from control, insulin-resistant, or type 2 diabetic patient groups using real-time PCR and did not find any correlation with insulin resistance. Savage and colleagues [57] were unable to detect resistin in lean patients but found increased levels of resistin mRNA in subcutaneous fat of morbidly obese individuals. However, similar to the results reported by Nagaev and Smith [56], resistin mRNA levels were detected in only 4 of 14 subjects in freshly isolated adipocytes and were not correlated with body mass index [57]. Savage and colleagues [57] also found that 24 h treatment of freshly isolated mononuclear cells with PPAR $\gamma$  agonists failed to have any effect on resistin levels, in contrast to the effects in mice and those reported by Patel et al. [58]. Furthermore, Savage et al. [57] found that a patient with clinically severe insulin resistance due to a mutation in PPAR $\gamma$  has no detectable resistin in subcutaneous fat, arguing that insulin resistance does not require elevation in resistin in humans [57]. Janke et al. [59] found that resistin is highly expressed in *pre*-adipocytes isolated from plastic surgery patients, and that resistin expression decreased during adipogenesis, contrary to the results in mice. Janke et al. [59] failed to find any correlation between adipose tissue resistin gene expression and body weight or insulin sensitivity. Several groups have investigated whether human resistin, as with mouse resistin, is increased in visceral adipose depots. Both Savage et al. [57] and McTernan et al. [60] failed to see a difference between resistin expression in subcutaneous and omental abdominal adipose tissue, although McTernan et al. [60, 61] detected 4.2-fold higher resistin expression [60] and protein [61] than in peripheral fat depots in thigh and breast.

## Genetic studies

As a common disease whose cause is multifactorial, it is unlikely that single gene mutations would account for more than a small percentage of type 2 diabetic cases. However, variants

of many genes, for example, insulin receptor [62], PPAR $\gamma$  [63, 64], and calpain-10 [65], have been associated with type 2 diabetes. Several groups have investigated whether the human resistin locus is associated with increased susceptibility to diabetes or obesity. Two studies have found an association with obesity but not with type 2 diabetes. In a study of nondiabetic population of Quebec, Canada, two single-nucleotide polymorphisms (SNPs) associated with the 5' flanking (promoter) variants were associated with body mass index [66]. Examination of the same variants in a Scandinavian population did not show this association, and neither population had any association with diabetes [66]. Another study conducted among whites in Boston, Mass., USA, found eight SNPs in the 5' flanking and intronic regions of the resistin gene [67]. This study failed to identify association with type 2 diabetes but did identify a SNP associated with obesity [67].

Two studies found associations of the resistin gene with changes in insulin sensitivity but not obesity. Pizzuti et al. [68] examined an Italian population of nondiabetics and found that an allele of an ATG triplet repeat in the 3' untranslated region is associated with lower fasting insulin, insulin resistance index, and serum triglycerides, suggesting a higher insulin sensitivity. Wang et al. [69] identified additional resistin SNPs which also were not associated with type 2 diabetes, but the SNP in the promoter region was a determinant of insulin sensitivity index. Finally, several studies did not find an association of resistin with either diabetes or obesity. Sentinelli et al. [70] discovered no sequence variants in the coding sequence, and a mutation in the 3' untranslated region was not associated with either diabetes or obesity. In a study of a Japanese population the identification of three intronic SNPs in 24 patients failed to identify any association with type 2 diabetes compared to controls [62]. Cao and Hegele [71] also failed to identify any resistin mutations but did isolate two SNPs to be used for future studies. At this time there is no consensus on the association of the human resistin locus with either diabetes or obesity.

## **Concluding remarks**

The discovery of resistin as a novel secreted factor regulated by a variety of hormones, drugs, and physiological states adds an intriguing molecule to the exciting family of molecules secreted by the adipocyte. Critical questions now need to be addressed: What is the role of resistin in normal physiology? What are target tissues for resistin? Does it have a receptor? Answers to these questions await the development and analysis of animal model systems

designed to alter resistin levels. These include ablating resistin expression by gene targeting, overexpressing resistin via transgenic approaches, and improved assays for detection and quantitation of resistin protein levels.

The fact that resistin is regulated by so many substances suggests that resistin plays an important physiological role. Investigations so far have focused on the effect of resistin on insulin sensitivity. A recently published study by Rajala colleagues [72] may help to clarify this contentious area. Using rats clamped at physiologically hyperinsulinemic concentrations, the authors found that acute infusion of resistin protein causes hepatic insulin resistance, primarily by causing the liver to fail to suppress glucose production despite the high insulin levels. This result is consistent with the glucose intolerance originally described in Stepan et al. [23] and suggests a possible mechanism for resistin action. However, in these rats no defect in the ability of peripheral tissues to take up glucose was observed [72]. Somewhat surprisingly, infusion of RELM $\beta$  protein had the same effects as resistin [72]. Future studies investigating the cellular mechanism of hepatic insulin resistance following resistin administration, and the effect of acute vs. chronic treatment with resistin will be critical in understanding how resistin influences insulin sensitivity. Other fundamental questions regarding resistin and RELM biology remain unanswered. Does resistin have its own receptor? If so, how does it crosstalk with insulin signaling pathways? Does resistin directly antagonize insulin action? How do the RELMS contribute to insulin resistance?

It has been suggested that resistin has a role in inflammatory conditions since it is expressed in human macrophages [56, 57], and RELM $\alpha$ /FIZZ1 is upregulated by inflammation in the mouse. However, the finding that resistin is downregulated by inflammatory mediators such as TNF $\alpha$  and LPS indicates either a divergence in mouse and human biology or argues against a role for resistin in inflammation. Molecular studies of resistin are already underway. In the mouse, for example, promoter analysis failed to identify any PPAR $\gamma$  binding sites within 6.2 kb of the transcriptional start site; the specificity of resistin expression is mediated through C/EBP $\alpha$  [39, 73]. Further understanding of molecular regulators of resistin may provide additional clues to its function and biology. It seems likely that resistin evolved for a purpose other than antagonism of insulin action. According to the “thrifty gene” hypothesis, humans have evolved under selective pressure favoring the ability to store energy and survive periodic starvation [10]. Resistin’s regulatory patterns in fasting and refeeding paradigms mirror that of leptin, which has been shown to be critical in the physiological response to long-term fasting [74]. This suggests that resistin is a candidate thrifty gene and may also

have a role in the starvation response. Perhaps under periods of prolonged fasting, where blood glucose levels fall, resistin blunting of insulin action is necessary or favors fatty acid substrate utilization. Resistin has also been proposed by Kim and colleagues [28] to block adipose tissue formation. Suppressing adipogenesis could contribute to insulin resistance by reducing the production of freshly differentiated, insulin-sensitive adipocytes. Suppressing adipogenesis could also provide adipose tissue a mechanism to maintain a pool of preadipocytes. Resistin has recently been suggested to be expressed at low levels in the pituitary and hypothalamus [75]. This and future studies will likely open the possibility for the involvement of resistin in entirely new and interesting pathways that will help to define its role.

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**Table 1.** Regulation of murine resistin gene expression

	Model system	References
Suppressors		
Thiazolidinediones	3T3-L1s, mice, rats	23, 24, 37, 38, 39, 40, 41
Obesity	Mice	20, 27, 42, 43, 47
Insulin	3T3-L1s	37, 38
Tumor necrosis factor $\alpha$	3T3-L1s	27, 38, 52
Isoproterenol	3T3-L1s	55
Epinephrine	3T3-L1s	38
Somatotrophin	3T3-L1s	38
Fasting	Mice	23, 27, 28
Lipopolysaccharide	Mice	27
Inducers		
Thiazolidinediones	Mice	42, 43
Glucose	3T3-L1s, rats	27, 38
Insulin	Mice	28
Dexamethasone	3T3-L1s, mice	37, 38
Hyperprolactinemia	Mice	53
Testosterone	Mice	53
Growth hormone	SDR rats	54
Refeeding	Mice	23, 27, 28
Lipopolysaccharide	3T3-L1s, rats	41
No effect		
Interleukin-6	3T3-L1s	27
Lipopolysaccharide	3T3-L1s, mice	27
Angiotensin-2	3T3-L1s	52
Growth hormone	3T3-L1s	52
$\beta$ -Agonists	3T3-L1s, mice	37, 40